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SOME METEOROLOGICAL FACTORS AFFECTING THE DISTRIBUTION OF FROST DAMAGE TO FRUIT TREES. I

By C. E. CORNFORD

INTRODUCTION.*

THE Agricultural Section of the Conference of Empire Meteorologists, held in London in 1929, recommended that the effect of varying weather conditions on the growth and cropping of fruit trees, especially spring frosts, should be studied. The severe frosts of the middle of May 1935 emphasized the importance of this recommendation, and at a Conference on Frost Damage, held at East Malling Research Station on September 11th, 1935, fruit-growers passed a resolution requesting the Ministry of Agriculture to provide facilities for a scientific investigation of the cause and prevention of frost damage to fruit and other crops. A programme of research work was drawn up suggesting that the nature of katabatic winds should first be investigated. The Ministry of Agriculture gave a grant to enable the writer to begin the research, with headquarters at East Malling Research Station, and this paper is a report of the preliminary work.

In 1924 the Ministry of Agriculture began a scheme for the collection and correlation of crop-weather data. Observations were made on certain horticultural crops, including apples, plums, black currants and peas. Irwin (11) has reported on that part of this scheme dealing with the effect of weather on the growth rates and dates of flowering of Bramley's Seedling and Worcester Pearmain apples, and Hoblyn (10) on the time of blossoming and on fruit set and "June drop". More detailed work on the effect of weather conditions in fruit plantations was carried out by Amos and others (1) in relation to incompatability; by Moore (13) in regard to the incidence of apple Scab; and by Srivastava (18) concerning the setting of pear blossom. After the severe frosts of the middle of May 1935, attempts were made to survey the damage done to fruit trees, and to explain how some orchards escaped it while others did not. The results of a survey, in which the writer took part, were reported by Bane (3),

* In this paper: (1) all observations apply to the topographical and climatic conditions of Kent and Hampshire; (2) unless otherwise stated, all thermometers were three feet above the ground, and were fitted with an anti-radiation shield; all temperatures are in Fahrenheit degrees; (3) unless otherwise stated, all night weather is that in which radiation of heat is taking place and air temperature is, on the whole, falling; (4) all times mentioned are civil times.

who discussed the problems involved in explaining the distribution of the damage. Other surveys were made by Spinks (17) and by Rothe (16).

This work showed that there is a reasonable prospect of determining in which fields crops are most likely to suffer from frost damage, and hence of solving certain problems connected with the choice of a site for a fruit farm. It also suggested other measures for reducing the danger from spring frosts, thus enabling existing methods, such as orchard heating, to be more usefully employed.

PREVIOUS LITERATURE.

General. Much has been written on the prevention of frost damage to fruit blossom. The writer has collected references to 600 papers and books on the subject. Many of these he has read, and summarized, but the summary is too extensive for inclusion here. The literature may be divided into two main groups: (i) American publications, dealing mostly with the practice of orchard heating, and (ii) European literature, dealing mainly with the science of micro-climatology.

American experience is, perhaps, best completely summarized in the booklet by Young (20) which explains, among other things, the meteorological fact on which the practice of orchard heating is based. That is, that on a calm, frosty night, the air temperature gradient in the first few hundred feet above the ground is inverted. In other words, the air temperature rises with increasing height up to that of several hundred feet. The depth of the inversion layer varies from night to night ; its upper limit is called the ceiling. Hot air rising from orchard heaters in a calm atmosphere does not rise indefinitely, but only to a level at which its temperature is identical with that of the air surrounding it. The air at this level is displaced and forced downward by other columns of hot air rising from the heaters. For every up-current there must be a corresponding down-current. In this way the air of the inversion layer circulates, and its temperature is raised, or at least prevented from falling to temperatures similar to those outside the heated area.

The essence of the European literature on micro-climatology is contained in the book entitled "Spring Frosts", by Day and Peace (8). The principal meteorological fact is that the ground may vary in its capacity to radiate heat and to cool the layer of air in the first few feet above it. Thus, on certain radiation nights, though perhaps not on all, the temperature conditions over each separate crop are determined to a large extent by that crop. Each crop produces its own micro-climate. To those interested in the prevention of frost damage the crops that give rise to the coldest and warmest air are most important. As shown by experiments reported in the present paper, the difference between the temperature of the air in a "warm" crop and that in a "cold" crop may

easily be as large as 6° F. Of the crops that give rise to relatively cold air on a radiation night, grassland is the most important. Bare soil gives rise to relatively warm air ; so also does a wood in which the leaf-canopy is complete. The reason why grass may give rise to relatively cold, and bare soil to relatively warm air on a radiation night, is as follows: The grass stems are poor conductors of heat and tend to prevent heat from the soil passing up them to the leaves to replace heat lost by them by radiation. The result is that the grass blades are greatly cooled, and they cool the air in contact with them. The heat lost from the surface of bare soil, on the other hand, is readily replaced by heat from below. Consequently the surface does not cool so much as grass does, with the result that the air above bare soil is warmer than that above grass.

In a wood in which the leaves form a canopy, but not perhaps in other types of wood, heat is radiated only from the upper surface of the leaf-canopy. The air in contact with the cooled leaves sinks among the trunks, which are at a relatively higher temperature and so raise the temperature of the cooled air, causing it to be several degrees warmer than air outside the wood in, for example, a meadow.

Frost pockets and frost plains. One consequence of the study of micro-climate is the necessity to distinguish between frost pockets and frost plains. A frost pocket is generally thought of as a valley in which cold air collects by gravitation. A frost plain is a field of grass, or other low crop capable of functioning in the same way as grass with respect to the radiation of heat, giving rise to a layer of relatively cold air above it.

Ramdas (15) and his associates working in India have found that on radiation nights the temperature of a bare soil surface is higher than that of the air in contact with it. Thus it seems unlikely that the soil, under these conditions, cools the air, as is often stated. In order to explain this, Ramdas has put forward the view that the air itself in the first one or two feet above the ground may cool by radiation.

Katabatic winds. The word katabatic means "down-going". A local wind is called katabatic if it is caused by the gravitation of cold air from high ground (2). Katabatic winds are also called downhill winds (5) and valley winds (14).

In 1824 Daniell (7) stated that as a result of radiation on the sides of low hills, "the air, which is condensed by the cold, rolls down and lodges at their feet". Other later authors have stated essentially the same thing (19, 4).

Marvin (12) drew attention to the fact that when air moves down a slope, its pressure, density and temperature increase. The temperature increases about 1·6° F. for every 300 feet decrease in height, provided that there has been no loss or gain of heat from other sources.

Newnham (14) stated that it is the difference in temperature between that

of the air near the ground on the slopes of the hills and that of the free air at the same level, which is the first cause of a valley wind, and that it matters very little what the temperature may be at the bottom of the valley.

Heywood (9) made observations in a valley (apparently devoid of woods or hedges) about one mile long and half a mile broad, the bottom of which sloped from an altitude of 600 feet to one of 400 feet. At the bottom of the lower end of the valley a Dines pressure-tube anemometer and a Baxendell direction recorder were set up, the vane being 15 feet above the ground. The readings of these instruments were compared with those of a standard Dines and Baxendell anemograph, working off a 42-foot mast standing on level high ground 2½ miles east-north-east of the instruments in the valley. The object of the investigation was to determine the velocity of katabatic winds in moderately hilly country and the conditions which give rise to them. The records of anemographs at the two stations were compared and it was found that a katabatic breeze blew down the valley on 29 out of 51 nights when, for several consecutive hours, there was a calm or a light wind in the open, and a clear sky. The katabatic effect was not specially frequent at any particular season of the year. The speed of the katabatic wind was usually very small, rarely exceeding 2¼ miles per hour. Both direction and velocity curves of the katabatic wind were narrow and regular, in striking contrast to the very gusty and irregular curves recorded during the daytime in the valley. While a katabatic wind was blowing in the valley, wind velocity in the open never exceeded 11 miles per hour.

The vertical temperature gradient in the surface layer of the atmosphere was measured by a temperature-gradient recorder at the higher station, and it was observed that while a katabatic wind was blowing there was always an inversion of temperature gradient above the high ground, usually of 1° to 5° F. between 100 feet and 4 feet. A katabatic wind was occasionally observed at the same time as a very large inversion of more than 5° F. between 100 and 4 feet. Such large inversions often reduced the katabatic wind to a dead calm. On several nights a katabatic flow started in the evening, but dropped to a calm at about the time when an inversion of over 5° F. was built up on the high ground. Heywood concluded that the cessation of the katabatic current while the conditions were still apparently favourable was due to the accumulation of cold air in the hollows.

With regard to the temperature distribution in a body of katabatic air, Young and Baughman (21) make the important statement: "Although relatively cold air drains slowly down even slightly sloping ground, its movement is so sluggish that the air which replaces it cools rapidly enough, through contact with the colder ground, to make the net effect of the drainage process almost negligible." They do not, however, mention any experimental evidence in support of the statement.

INSTRUMENTS.

A variety of instruments was used in this research work. Some of them were of standard pattern and are sufficiently well known as not to require description here. The little-known anti-radiation screen, used on all thermometers, and the instrument devised by the writer for recording the speed, direction and temperature of the katabatic winds were described by him in the Quarterly Journal of the Royal Meteorological Society for October 1938.

STUDY OF KATABATIC WINDS.

Certain features of katabatic winds which have apparently not been reported before, became evident in the first night on which they were investigated.

On the clear evening of July 26th, 1936, there was a "calm" in the Medway Valley near Aylesford. The word "calm" does not mean that the air was absolutely still, but that the wind moved at less than a walking pace, i.e. 2 or 3 miles an hour. Winds of this speed are not normally noticed, but their existence may easily be demonstrated by the use of a lighted candle,

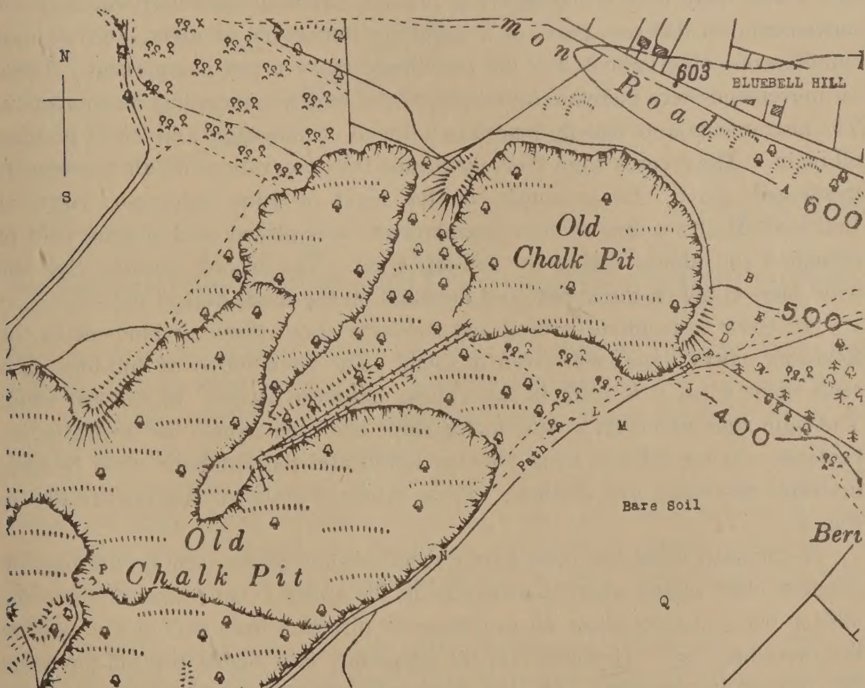

Fig. 1.
Contour map of the 1 : 3 slope at Bluebell Hill. The area shown measures 450 by 500 yards. (Profile in Fig. 5.)
Based upon the Ordnance Survey Map, with the sanction of the Controller of H.M. Stationery Office.

smoke, hanging threads of Angora wool or thistledown. Although the speed of air currents in the valley bottom was of this order on the night in question, at place A, Fig. 1, on the 600 feet contour of Bluebell Hill, about 2 miles away, a much stronger wind of 5 to 10 miles per hour was blowing. It came from the S.S.W., and in a meadow facing S.S.W. on the hilltop the wind blew uphill. However, smoke from cottage chimneys in the valley showed that at lower altitudes the air was moving in a contrary direction, i.e. downhill. Walking downhill in the meadow facing south the force of the south wind grew less, until a level was reached at place D, Fig. 1, where no definite air movements could be detected by such sensitive means as hanging threads or pieces of thistledown. Below this calm zone the air moved steadily downhill at about a walking pace, i.e. at such speed that the existence of the air current might easily be overlooked. This downhill wind must have been a katabatic wind, because it blew in a direction opposite to that of the main wind.

These air movements were at first investigated by using hanging threads of Angora wool, thistledown and smoke. Such methods suffer from the limitation that unless more than one observer is present, simultaneous observations of air movements on different parts of a slope are impossible at night, and without simultaneous observations it is not possible to make a just comparison. These air movements were therefore investigated later with five recording anemometers. The instruments were usually placed in a line at evening time and left in position all night. The records from them confirmed the observations on air movements mentioned above. For example, on the night of June 23rd-24th, 1937, at Leckford, Hants, a line of four anemometers was placed in a sloping field of ploughed soil almost facing the hill-top wind. The records showed that the wind blew uphill on the upper, and downhill on the lower part of the slope.

In order to facilitate writing and speaking about these air movements the following names have been given to them. The wind found on hill-tops on a clear night when it is calm in the valleys has been named the hill-top wind. The calm zone found on a wind-facing slope has been named the neutral zone, because in it the hill-top wind blowing uphill and the katabatic wind blowing downhill neutralize one another. These winds are shown diagramatically in Fig. 2.

The hill-top wind has been found at all seasons of the year on hill-tops on nineteen clear nights when it was calm in the valley. On fifteen of these the hill-top wind was not observed to change its direction from that of the wind of the preceding day. However, on the remaining four nights the hill-top wind did change its direction. On the night of August 6th-7th, 1937, at about 3 a.m., for example, the hill-top wind at Leckford changed from S. to N. Thus, when making observations on the influence of the direction of the hill-top wind on that of the katabatic wind it cannot be assumed that the hill-top wind

never changes its direction. This again emphasizes the need for making observations in the hill-top wind and in the katabatic wind simultaneously.

The detection of the hill-top wind suggests the possibility of using it for avoiding frost damage to orchards situated on relatively high land. In such places exposure to the hill-top wind seems likely to be beneficial, under certain conditions in severe frost, but it might be very detrimental to the orchard in other ways and at other times of the year.

The neutral zone is not always present in quite such a clear-cut form as that described above, in which it has been observed on five nights.

On the night of June 15th-16th, 1937, at Leckford, the hill-top wind was blowing from the N.W. on a slope facing W.N.W. Observations made both with the aid of smoke and a line of recording anemometers showed that the air in the neutral zone moved along the contours to the S.S.W., i.e. neither

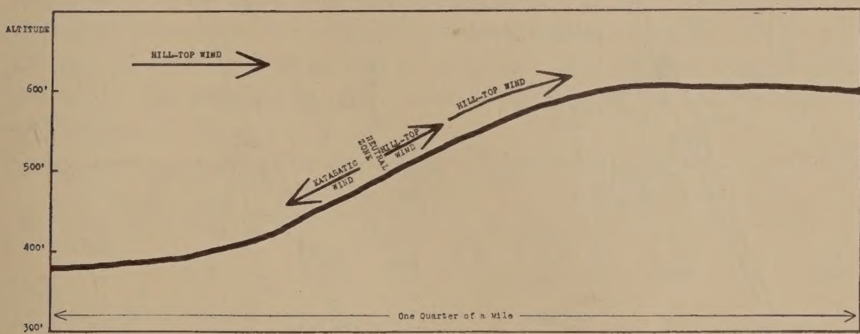

FIG. 2.

Section, drawn to scale, through Bluebell Hill, showing typical air movements on a radiation night when the hill-top wind blows directly upon the slope.

up- nor downhill. Other similar observations, made on different slopes and with the hill-top wind blowing in a different direction, showed the same thing, viz. that when the hill-top wind blows obliquely on the slope the air in the neutral zone moves along the contours.

Above the neutral zone, on the night of June 15th-16th, 1937, the air moved obliquely uphill, carried in the N.W. wind ; and below the neutral zone the air took a semi-circular path downhill. All these air currents are shown on the right of Fig. 3. Observations carried out on other nights confirmed these facts.

Katabatic winds in woods. In early experiments with smoke, on the steep slope of Bluebell Hill on the North Downs, it was noticed that the katabatic wind on the lower part of the slope often passed, with no perceptible loss of speed, into shrubs and bushes having branches reaching to the ground. The katabatic air seemed to move through the bushes as freely as in the open.

Closer investigation with the aid of smoke, threads and thistledown, showed that a katabatic wind was blowing from the lower edge of a wood, at a place K, Fig. 1, that covers the steep slope. This particular wood is perhaps more accurately described as a thicket. No canopy is formed by the tops of close-standing trees, but beech trees about 30 feet high are widely spaced, and between them there is a dense tangle of undergrowth reaching to the ground. On two consecutive nights (September 4th-6th, 1937) a line of five recording anemometers was placed on this slope, the highest instrument being in a field above the wood and fully exposed to an uphill wind. The lowest stood in a field below the

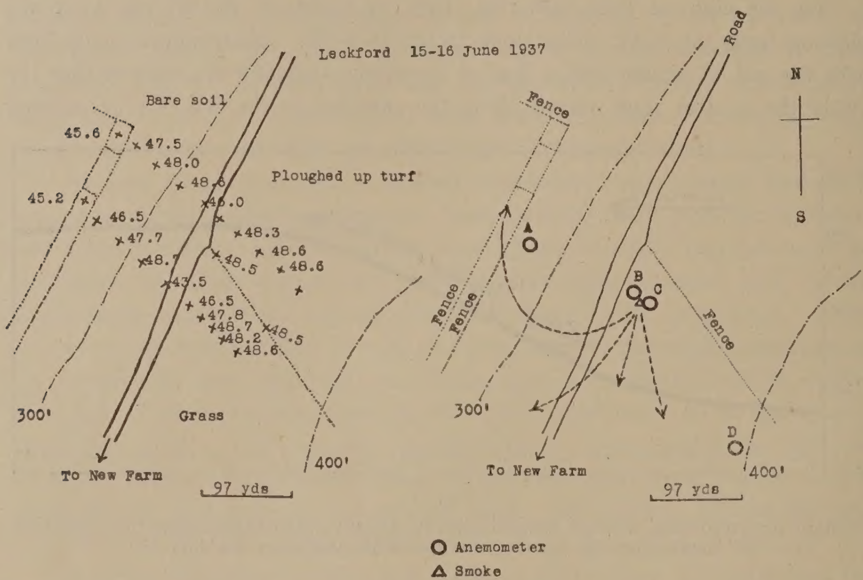

FIG. 3.

Map of slope at New Farm, Leckford, showing, on the right, positions of four anemometers on the night of 15-16 June, 1937, and paths taken by various parts of a smoke cloud liberated in the neutral zone at 10.00 p.m. on 15 June, 1937. The positions of thermometers are shown on the left with the minimum temperatures recorded. These are shown in graph form in Fig. 14.

wood, and the remaining three stood in the wood. The records of these instruments confirmed those obtained with smoke, threads and thistledown, and showed that in this particular wood the air movements were essentially the same as on a slope devoid of a wood. In the upper part of the wood the wind blew uphill, and in the lower part downhill ; between these two winds there was a neutral zone in which the air, although not absolutely still, moved in no definite direction. The highest anemometer was in an uphill wind all night, and the lowest in a downhill wind. The neutral zone did not always remain in the middle of the wood. On the night of September 4th-5th, 1937, for example,

it moved from the middle to the upper part of the wood and even into the field above.

Recording anemometers were also placed in a different type of wood on a steep slope facing the hill-top wind, at Leckford, on the night of July 7th-8th, 1937, and similar results were obtained.

These results show that vegetation which to the eye looks "solid", need offer little resistance to the motion of air moving at a low speed. This does not mean that all vegetation is equally pervious to air currents of low speed. For example, a beech hedge was found to be impervious to a katabatic wind, for it flowed over it. The wind blew at a speed of 50 to 100 feet per minute (1 m.p.h.) at 1 metre high over bare soil, in a direction almost at right angles to the hedge, which was 2 metres high and 1 metre thick. Two Ower, low-speed, anemometers were used, one being placed on the windward side of the hedge half a metre from it, the other in a similar position on the other side, and they were read simultaneously. The instruments were held at varying heights above the ground, and were kept parallel to the hedge. The following were the results obtained. The numbers 1·3, etc., refer to divisions of the scale on the Ower anemometer.

Time.	Leeward Side.				Windward side.			
	Height above ground in metres							
	½	1	1½	2	½	1	1½	2
7.7 p.m. ..	0·0				1·3			
7.10 ,, ..		0·0				1·5		
7.13 ,, ..			0·0				1·0	
7.15 ,, ..				0·0				2·3

These records show that a wind blew on the hedge but did not penetrate it. No air movement was found in the middle of the hedge when there was air movement on its windward side. Smoke observations showed that the air flowed over the hedge.

Similar effects were observed on the evening of September 29th, 1937, when a photograph (Fig. 4, Plate I) was obtained showing what happens when a katabatic wind meets a six foot stone wall ; it passed over the wall and was not held up by it. Thus, these few observations tend to show that it is difficult to influence the movements of wind of low velocity by means of shelter-belts, wind-breaks and walls, but this of course must not be taken to imply that shelter-belts, wind-breaks and walls cannot be of service in the prevention of frost damage. In the next section of this paper it will be shown that the movement of a katabatic wind is by no means always directly related to its temperature, and that it cannot be assumed that every katabatic wind that enters a field brings relatively cold air with it.

Air movements on the leeward side of a steep slope. On a slope facing the hill-top wind, the katabatic wind and hill-top wind blow in contrary directions, making it easy to recognize the katabatic wind simply on the evidence of its direction. For this reason most of the present work has been carried out on wind-facing slopes, where there is certainty as to whether a katabatic wind is present or not.

On one night, March 22nd, 1937, when katabatic winds were blowing on the lower parts of slopes facing the north hill-top wind, investigations were carried out with the aid of smoke on the leeward side of a steep slope facing

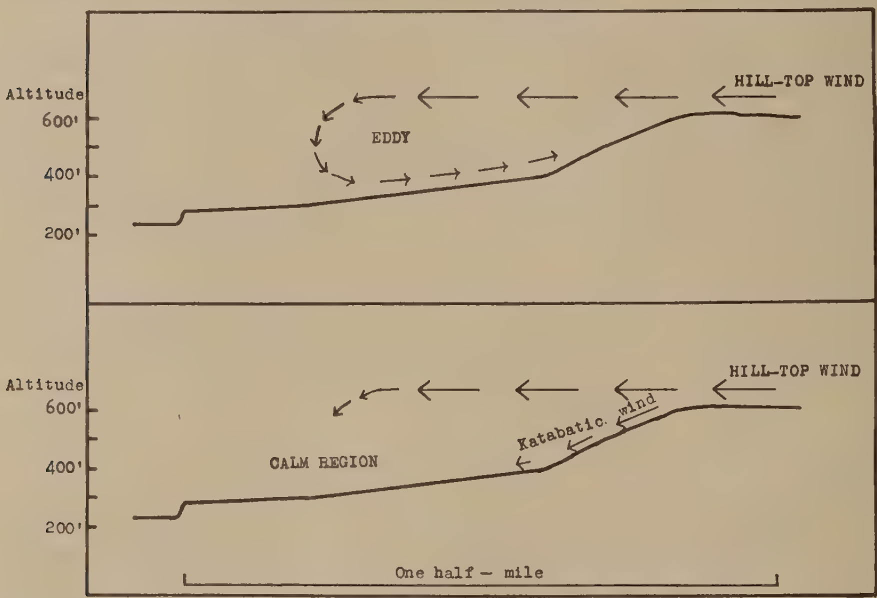

FIG. 5.

Top.—Diagram of section of Bluebell Hill, drawn to scale, showing an eddy on the leeward side of the hill under non-radiation conditions.

Bottom.—As above, showing influence of katabatic winds on the eddy as observed on the night of March 22nd-23rd, 1937.

south. It was found that the air to a depth of several hundred feet at the foot of the steepest part of the slope (at Q, Fig. 1) was remarkably calm. This may be explained by assuming that an eddy was formed in the lee of the hill by the hill-top wind. The return current of this eddy, blowing uphill, and the katabatic wind, blowing downhill, on this occasion counterbalanced one another (see Fig. 5).

The records of anemometers standing in the bottom of the valley at Leckford, shown in Fig. 6, demonstrate that on several nights when the hill-top

wind blew in the same direction as the katabatic wind, the latter moved no faster than when the hill-top wind blew in the contrary direction to that of the katabatic wind. This effect was clearly seen on the night of August 6th-7th, 1937, when the katabatic wind blew steadily straight down the valley while the hill-top wind blew from almost all points of the compass.

FIG. 6.

Contour map of the 1 : 3 slope at Leckford, Hampshire, showing crops and minimum temperatures recorded on the night of July 7th-8th, 1937.

Smoke observations made on only a few nights in the Leckford valley, indicated that under radiation conditions the katabatic wind in the valley bottom was about 15 feet deep, and when the hill-top wind blew up the valley in a contrary direction to that of the katabatic wind, the air above the katabatic wind moved at a walking pace up the valley. These air movements are shown by smoke in Fig. 7 (Plate I), and their effect on the hot air from orchard heaters would be to tend to carry much of it down the valley and the remainder up the valley, an effect which was actually observed with smoke.

FACTORS INFLUENCING AIR TEMPERATURE ON A RADIATION NIGHT.

In the previous section the conditions affecting the direction of air movements in the first 10 or 20 feet above the soil on radiation nights were dealt with. The temperature of this air has also been investigated, chiefly with the help of minimum thermometers placed 3 feet above the soil. Thermographs were also used.

The first experiment was designed to ascertain the effect of altitude on air temperature. A line of five thermometers was placed across one valley at Wateringbury, Kent, and a line of five thermographs across another valley at Aylesford, Kent. After each radiation night in January and February 1937 the minimum temperatures were noted. It was found that on certain nights the lowest places were coldest and the temperatures increased with altitude, as expected. On one other night, however, when the temperatures were all at or below freezing point, showing that radiation had taken place, the lowest places were warmest and the temperature decreased with altitude. It is proposed to refer to the latter type of temperature distribution as "anomalous". The temperature records showing both these phenomena on typical nights are shown in Fig. 8.

On five of nine radiation nights, from January 12th to February 12th, 1937, inclusive, anomalous temperature distribution was observed in varying degrees of clarity. Hence it seems to be common, and if so might help to explain certain facts relating to the distribution of frost damage, which is not always most severe in valley bottoms.

In the next experiment, of February-March 1937, five thermometers were placed at the bottom of an inland part of the Medway valley several miles long. The minimum figures showed that air temperatures did not decrease as altitude decreased. The temperatures at the various points bore a remarkably constant relation to each other. For example, on each radiation night the temperature at Halling was always the highest, and that at Aylesford, higher up the river, always the lowest. Hence it was evident that a constant factor or group of factors was determining the minimum temperature at each point.

In the two previous experiments it had been noticed that the thermometer at Aylesford always recorded a minimum temperature far lower than any other. The reason for this was sought by placing minimum thermometers in the fields near it. In this way records were obtained of the temperatures in certain fields of two neighbouring parishes, Aylesford and Ditton. This land is generally

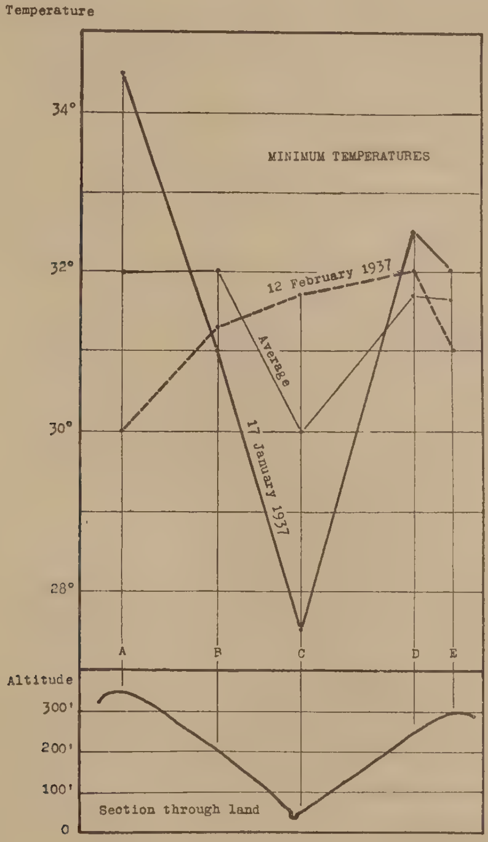

FIG. 8.

Graph showing relation of minimum temperatures to altitude in a river valley. A section of the valley is shown at the bottom. The distance from A to E is 1·5 miles. Vertical lines connect temperatures with positions on the slope.

low-lying, but is undulating. There are not many orchards in this district, and the few there are do not all crown the undulations ; some are on the lowest land.

The records from these experiments all showed that altitude was not the only factor affecting air temperature. Frequently on one night a place at a low

altitude was warmer than a place at a higher altitude, and on the same night, in another locality, the reverse was recorded.

To take a few examples. A combined frost pocket and frost plain near Ditton Church, at a higher altitude (50 feet) than Aylesford (20 feet), had nevertheless a minimum temperature about 2° F. lower than the latter on a frosty night. These two places are one mile apart.

Another example was found in which two places one twelfth of a mile apart and having altitudes of about 30 feet and 40 feet respectively, had minimum temperatures differing by two degrees in the same frost, the lower place being the warmer.

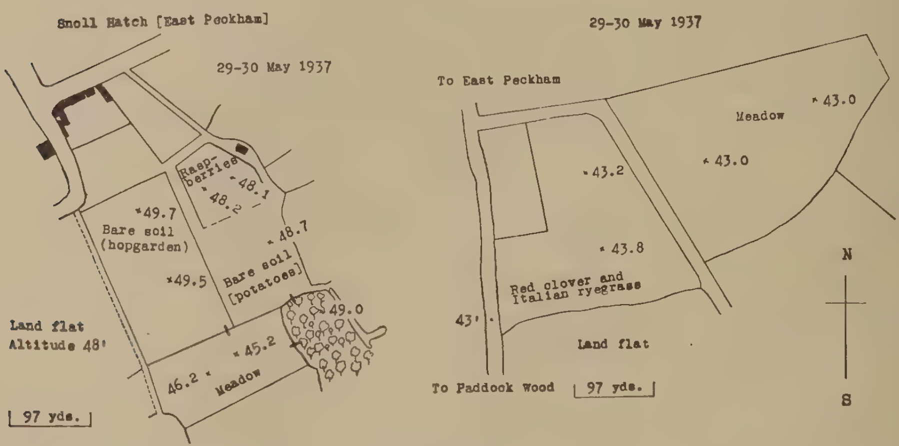

FIG. 9.

Maps of fields at East Peckham showing, with crosses, positions of thermometers, and crops corresponding to those shown in Fig. 10. The numbers against the crosses are minimum temperatures recorded on the night of 29-30 May 1937. The three thick short lines connecting the hopgarden with the meadow, the meadow with the wood, and the wood with the potato field, represent lines of five thermometers shown in Fig. 12.

Yet another example is afforded by comparing Aylesford Sand Pit, flat-bottomed, devoid of grass, twenty acres in extent, partly cultivated, and surrounded by 25 foot cliffs, having its bottom at an altitude of about 25 feet, and East Malling Research Station, altitude 100 feet. On the same frosty night both places had the same minimum temperature. These two places are 1·5 miles apart.

Hence it may be concluded that isotherms at three feet above the ground on a frosty night do not always follow the contours.

It was noticed that in a large meadow at Aylesford, where there was relatively high land in the middle and relatively low land round the edge, the air temperature varied with the altitude. Thus, over uniform vegetation, air

temperatures bear a direct relation to altitude in certain places on some radiation nights. Also, it was noticed that the three coldest places were all in or near meadows. It was thus found that the vegetation in certain fields determines to a large extent the temperature of the air above them on a radiation night. The thermometer at Aylesford which had recorded remarkably low minima was in a garden a few yards from the lower edge of a large pasture on a slope. The

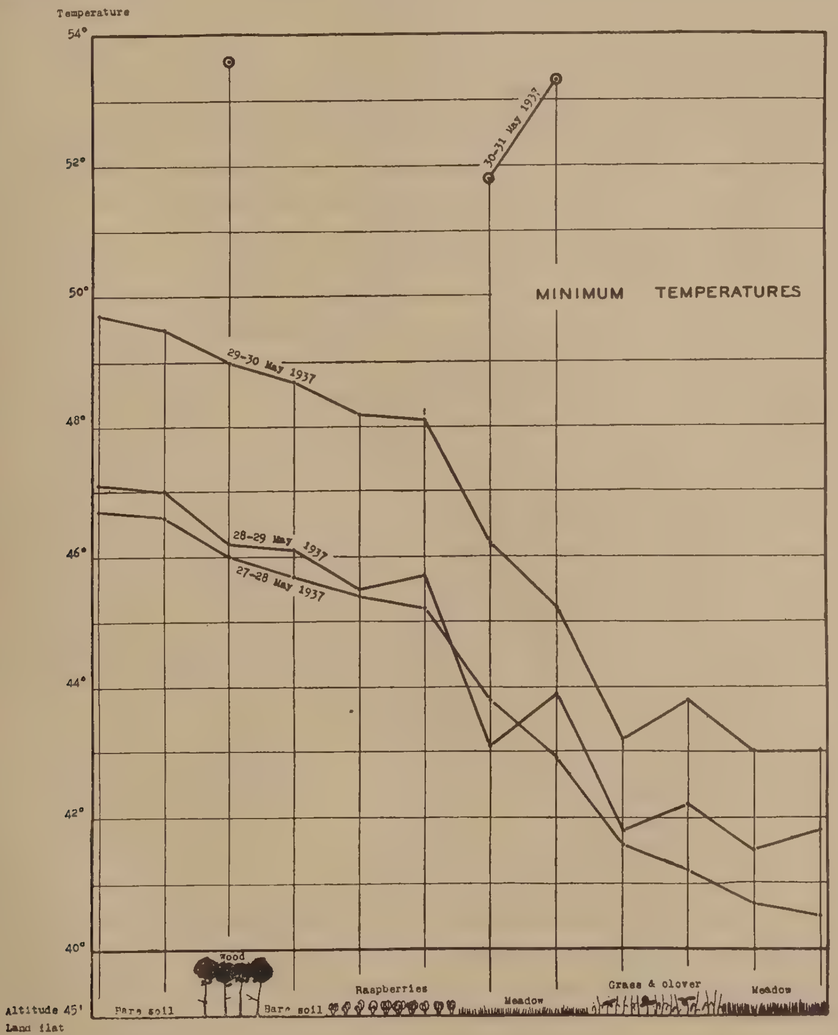

FIG. 10.

Graph of minimum temperatures, arranged in descending order, recorded over the crops shown at the bottom on three consecutive radiation nights in May 1937, at East Peckham.

experiments had shown that air over a pasture is liable to reach the lowest temperature recorded on a frosty night, and this explained the low minima that had consistently been recorded near the edge of the pasture.

It was argued that if vegetation was a factor affecting air temperature, it would be desirable to determine the magnitude of its effect under conditions in Kent. With this in view experiments were carried out on level land at East Peckham, shown on the map, Fig. 9, Beltring and Yalding, on which, of course, the factor of altitude was eliminated. Thermometers were placed in the middle of fields growing various crops, including fruit.

The results of the first experiment are typical of later ones. The minimum temperatures of the night of May 27th-28th, 1937, are given in Fig. 10. They show that the lowest air temperatures occurred over meadows and a field of grass and clover. The highest air temperatures were found over bare soil and in a wood the canopy of which was complete. In a field of raspberries, air temperatures were intermediate between those in the meadows and those over bare soil. The difference between the highest and lowest temperature recorded was 6°. On the two following nights these differences were 5° and 6·5° respectively. These differences may be compared with those that occurred between Aylesford, altitude 40 feet, and Bluebell Hill, altitude 600 feet. The average difference for nine radiation nights was 6·2° F.

The East Peckham experiment showed therefore that differences in air temperature caused by vegetation may be of the same order as those caused by topography in this part of England.

On the night of May 30th-31st, 1937, two thermometers were placed in deep grass that reached a height of 3 feet in the middle of a standard apple orchard, and two others were placed in the next field where the soil was bare. The apple trees were in leaf, and formed a complete canopy. The minimum temperatures were as follows:—

Grassed apple orchard	53·2° F.
,, ,, ,, ...	53·2° F.
Bare soil	53·7° F.
,, ,, ...	53·5° F.

The four temperatures were practically the same, although on the same night a thermometer over grass exposed to the sky about 100 yards from the other four recorded a temperature of 51·3° F., i.e. 2° lower, showing that radiation took place. This experiment indicates that it is only exposed grass which is able to produce relatively cold air above it on a radiation night. The same effect was observed in other places on the nights of May 30th-31st, May 30th-June 1st, and June 4th-5th, 1937.

The results of the East Peckham experiment showed that the air over one meadow was always colder by about 3° F. than that over the other. As far as could be seen no difference existed in the soil and drainage in the two meadows, and it is thought that the effect was due to the fact that the grass in the warmer meadow was shorter and more irregular than that in the colder meadow, where the grass was taller, denser and uniform. This indicates that tall dense grass is more to be feared as a producer of cold air than short grass.

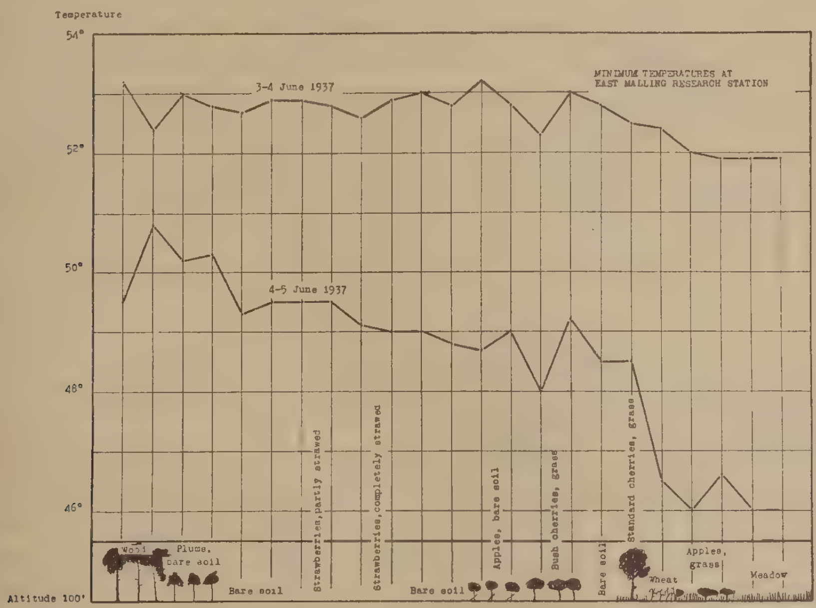

FIG. 11.

Graph showing minimum temperatures at 3 feet in various crops at East Malling Research Station, arranged in decreasing order. The records for the non-radiation night 3-4 June are given for comparison.

Minimum temperatures at 3 feet high, recorded in early June in various plots at East Malling Research Station, and near it, are presented in Fig. 11. The records show:—

(1) That the difference between the highest and lowest temperature on the night of June 4th-5th, recorded among fruit crops, was 4·2°, the highest temperature being practically the same as that recorded in a wood the canopy of which was complete, and the lowest being the same as that recorded in a meadow.

(2) That as great a variation occurred among air temperatures in fruit crops as in other fields, indicating that a relatively great range of micro-climates exists among fruit crops.

On the last of the three nights during which the experiment was carried out at East Peckham, and when recording anemometers showed that almost a dead calm existed, an attempt was made to discover to what extent the relatively cold air over a meadow affected the temperature of the relatively warm air over an adjacent piece of bare soil. A line of five thermometers, two yards apart, was placed at right angles to a fence separating a meadow from a field in which the soil was bare. The middle thermometer of the five was placed on the fence. On the same night two other similar lines were set up, one stretching from a wood into a meadow and the other stretching from a wood into a field of bare soil (see Fig. 9). The results are shown in Fig. 12, from which it will be seen that there was a sharp change in air temperature near the line separating the meadow from the bare soil. However, the air over the meadow slightly cooled the air over the bare soil for a distance of at least four yards, because it was cooler over the bare soil near the meadow than in the middle of the bare soil, as the following figures show:—

Middle of bare soil	49·7° F.
,, ,, ,, ,, ...	49·5° F.
Four yards from edge of bare soil ..	46·7° F.
Two ,, ,, ,, ,, ,, ,, ..	46·7° F.

The records from the other two thermometer lines confirmed these conclusions, as also did records from similar experiments made at Yalding on the night of May 30th-31st, 1937. An experiment made on the night of June 6th, 1937, at Birling, Kent, where a field of wheat was surrounded by bare soil, was similar, except that a katabatic wind, moving at a speed of two or three miles per hour, was blowing over the ground. Under these conditions there was also a sharp change in air temperature at the line separating a wheat field from a field of bare soil, as shown in Fig. 13. The following figures illustrate the magnitude of the temperature differences recorded on this occasion, at 3 feet high:—

At A, Over bare soil, to windward of wheat ..	64·0° F.
At B, In the middle of the wheat	62·6° F.
At C, On the leeward edge of the wheat	62·0° F.
At D, Over bare soil, 80 paces to leeward of wheat	63·6° F.

These figures also illustrate a fact of importance to horticulture and one which apparently has not been noted before, namely that under certain conditions the temperature of a katabatic wind depends to a large extent on the crop over which it flows.

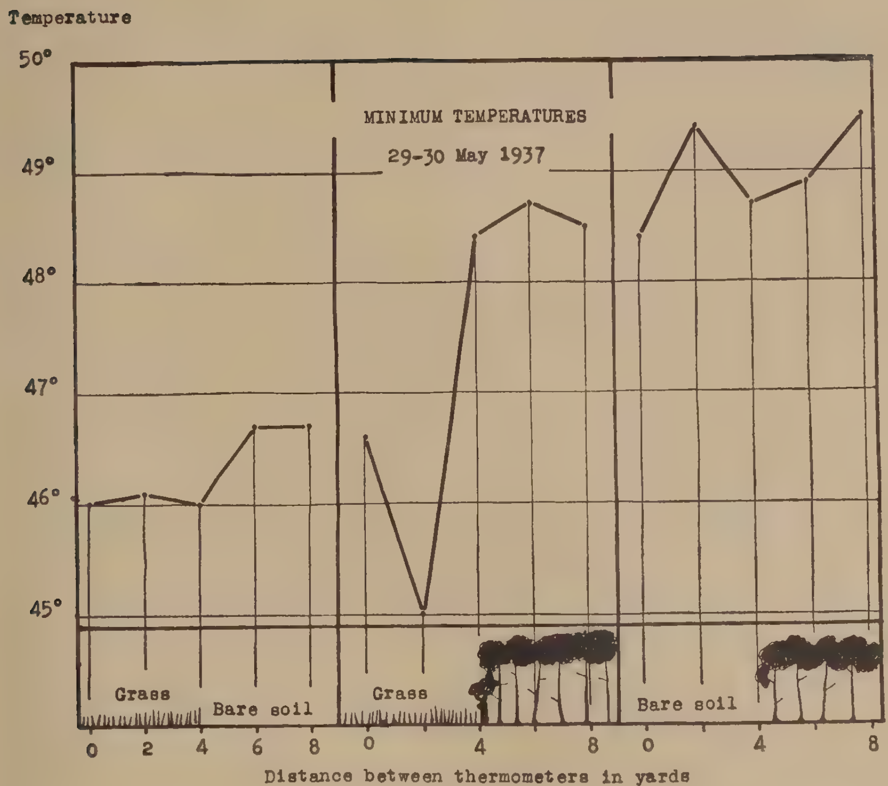

FIG. 12.

Graphs showing minimum temperatures recorded by three lines of five minimum thermometers placed near the edges of certain fields shown on the left of Fig. 9.

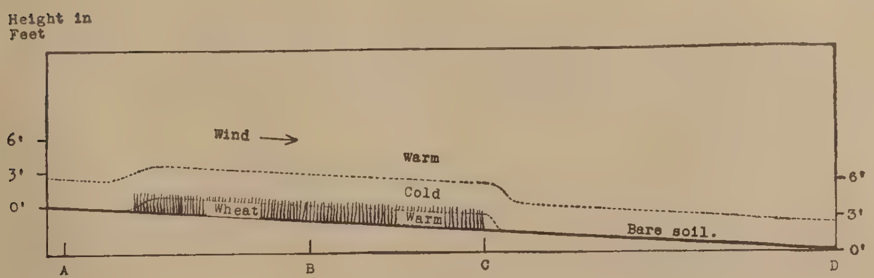

FIG. 13.

Diagram (not to scale) of sloping fields of bare soil and wheat, showing the temperature profiles over bare soil and wheat in a katabatic wind.

In the Birling experiment, the thermometer line was parallel to the direction of the katabatic wind, so that the air which passed the windward end of the thermometer line also passed the other thermometers. The temperature of this air fell about 2° F. while it was moving over the wheat, and rose again to almost its original figure when it again moved over bare soil. Other experiments also illustrate the same fact (see for example the data presented in the left half of Fig. 3, and in Fig. 14).

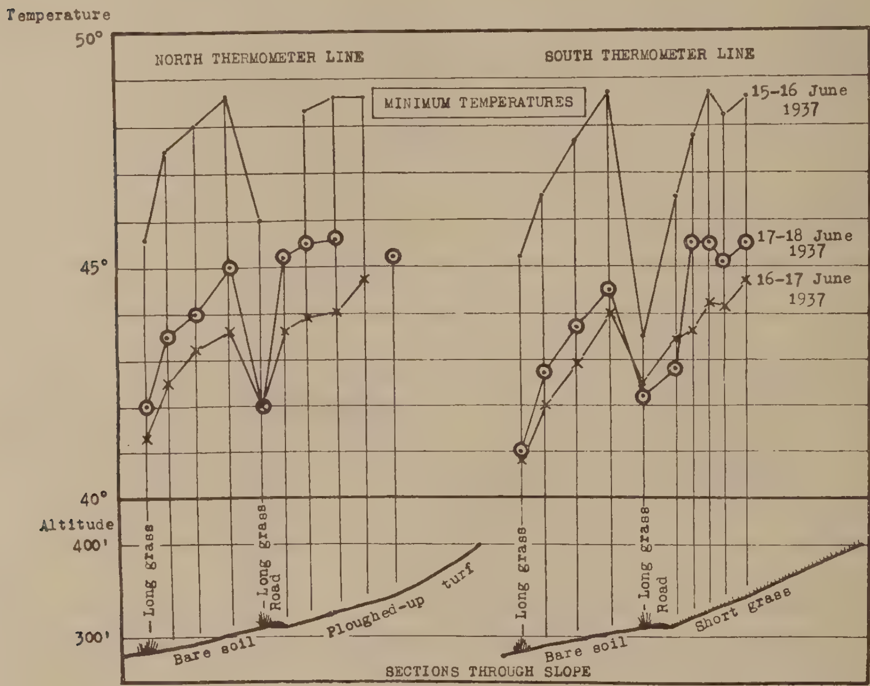

FIG. 14.

Graph of minimum temperatures, recorded on the slope shown in Fig. 3, on three consecutive nights, with sections of the slope drawn to scale. Vertical lines connect temperatures with the places where they were recorded.

Perhaps the best illustration is afforded by the experiment of July 7th-8th, 1937. This was carried out in the valley at Leckford in which, by good fortune, the higher part was in grass and the fields in the lower part were devoid of grass, as shown in Fig. 6. A line of eighteen thermometers was placed in the bottom of the valley, and others were placed on its sides. A recording anemometer was also placed in the valley bottom and others on the sides. The temperature records presented in Fig. 6 and in Fig. 15 show that the lowest temperatures were recorded over the highest part of the valley, that in grass.

In the lower part the temperatures were 3·5° higher, in some places, than over the grass in the higher part. This was so in spite of the fact, shown by the anemometer, that air moved down the valley from the colder to the warmer part, i.e. in such a direction as to tend to diminish the difference in air temperature between the two parts.

This experiment also shows that air temperature differences at 3 feet above the ground caused by vegetation can be large enough to offset those caused by topography.

That topographical influences alone can cause air temperature differences was mentioned above. The fact is generally accepted but it has apparently not

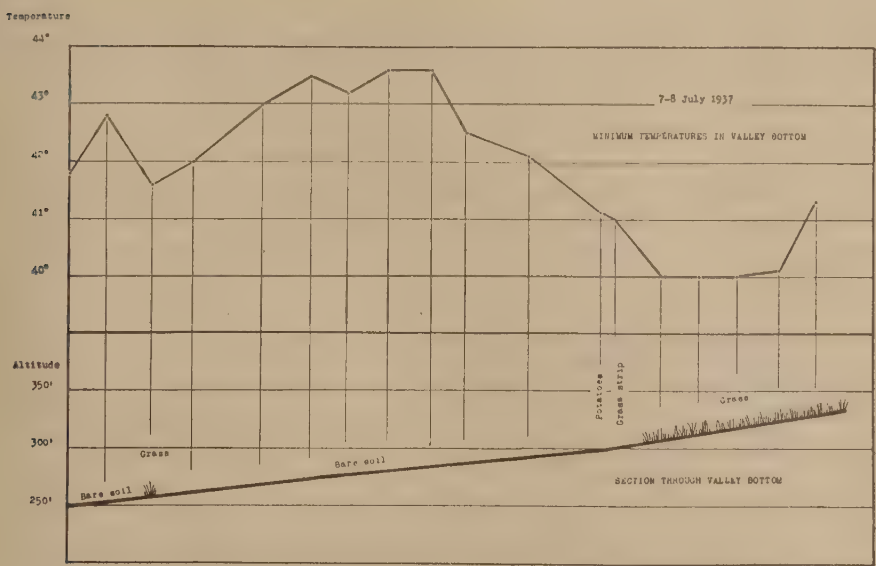

FIG. 15.

Graph of minimum temperatures recorded on a line of thermometers placed in the bottom of New Farm Valley, Leckford, with a diagram of a section through the valley bottom showing the vegetation. Vertical lines connect temperatures with the places where they were recorded.

been conclusively proved. In order to do this, temperature variation due to all other factors must be eliminated. On the night of April 3rd-4th, 1937, temperature variation due to differences in vegetation was eliminated when a line of ten thermometers was placed across a little valley in a meadow (see Fig. 16). Three similar experiments were carried out at Leckford. That of June 29th-30th, 1937, is an example, as shown in Fig. 17. The minimum figures indicate that over both grass and bare soil the temperatures varied with altitude, the lowest

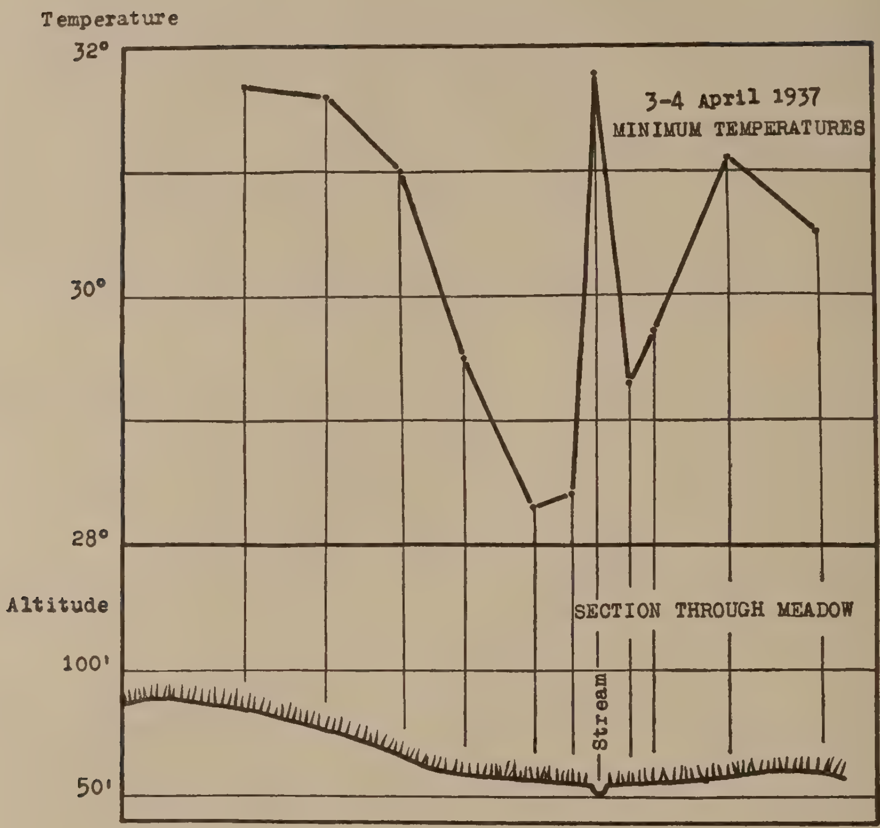

FIG. 16.

Graph of minimum temperatures recorded on a line of thermometers placed at 3 feet high in a valley in a meadow, with a diagram of a section through the valley. Vertical lines connect temperatures with the places where they were recorded.

places being the coldest. However, it must be remembered that anomalous temperature distribution (see Fig. 8) may reverse this condition and cause the lowest place to be the warmest although all temperatures may be below freezing point and consequently a danger to fruit blossom.

EXPERIMENTS WITH ORCHARD HEATERS.

An example of how the findings reported above can be applied in practice is afforded by the following account of experiments designed to discover the best method that could be applied to investigate the effectiveness of heaters in raising the temperature of the orchard air.

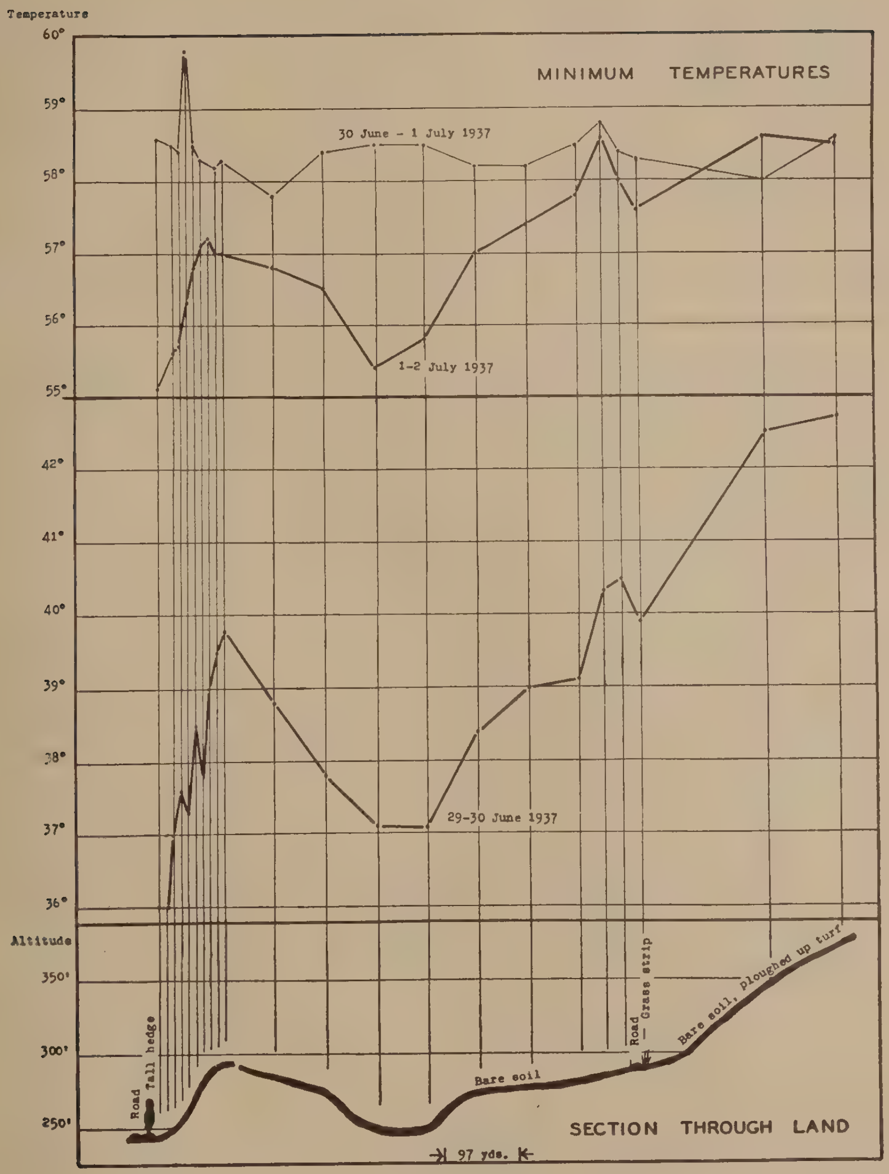

Fig. 17.

Graph similar to Fig. 16 showing variation of temperature with altitude over bare soil. For comparison, temperatures from a non-radiation night, 30 June-1 July, 1937, are given.

To obtain a comparison of the temperature of the air in the heated area with that outside it, two methods were employed:—

(1) Screened minimum thermometers were placed on a mast at heights of 3 inches, 10 feet and 20 feet in the middle of the heated area, and a similar control mast bearing thermometers was placed about 200 yards outside the heated area in the same orchard. The temperatures recorded on both masts were compared.

(2) The temperature recorded by a whirled unscreened thermometer, held at about 3 feet high on the leeward edge in the middle of the heated area, was compared with that recorded by a similar thermometer on the windward edge of the heated area in the same orchard.

Experiments were carried out on five nights, of which that of April 22nd-23rd, 1936, is typical. On this occasion the heated area was a one acre square of standard and bush apples and plums in the middle of an eighteen acre plantation of trees standing 12 feet apart both ways, situated on almost level ground, at East Malling Research Station. The trees in the heated area were about 20 feet high and the branches of adjacent trees were long enough to touch one another; the soil had been cultivated between the rows but not near the trunks of the trees, so that for 2 feet round the trees there was grass about 6 inches high, while on the remaining ground there was little grass and the soil was almost bare. Conditions were similar near the control mast, except that the trees were about 10 feet high and their spread was about 8 feet, so that more sunlight reached the ground, and grass grew more abundantly near the mast than in the heated area. These differences in the vegetation near the two masts had an important bearing on the experimental results. The acre was heated by sixty Harrington heaters, uniformly distributed, which were lighted at 11 p.m. and extinguished at 7 a.m. In addition to the thermometers mentioned above there were eight more on poles at 10 feet above the ground in the heated area and two thermographs at 50 feet high, one above the middle of the heated acre, and one on the control mast. The thermometers on the masts were read simultaneously at hourly intervals, from 2.10 a.m. onwards; those on the eight posts were read successively nine times in the six hours following midnight.

A comparison of temperatures made on the lines of method (1) described above showed:—

(a) that at 50 feet there was no observable difference between temperatures recorded in the heated and unheated areas, although as shown in Fig. 18, smoke from the heaters had enveloped the thermograph above the heated area.

(b) that at 20 feet it was colder in the heated than in the unheated area. At 6.10 a.m., for example, the temperatures recorded were:—at 20 feet, 200 yards from the heated area, 32·4° F.; at 20 feet, in the middle of the heated area, 30·9° F.; difference = 1·5° F.

(c) that at 10 feet the eight thermometers in the heated area, at any one time, recorded differences of from 2° to 4° F., depending on the position of the pole with respect to the nearest heater. On the whole however it appeared to be about 1° F. warmer at 10 feet in the heated area than at the same height about 200 yards away.

(d) that at 3 inches, temperatures were about 0·75° F. higher in the heated area than in the unheated area, the minima recorded at about 6 a.m. being:—heated area 28·1° F.; unheated 27·6° F.; difference = 0·5° F., both temperatures being below freezing point.

A comparison of temperatures made on the lines of method (2) described above showed that the difference between the temperature of the air on the leeward and the windward sides of the heated acre at a height of 3 feet above the ground and 24 feet from the central heater of the appropriate side was about 1·5° F. Readings taken at 2.30 a.m. were:—leeward side 33·5° F.; windward side 32·1° F.; difference = 1·4° F. On the night of May 23rd-24th, 1936, when the heaters were not lighted, the temperatures at 3 inches were higher in the heated area than outside it. Readings at 12.30 a.m. were:—heated area (heaters out) 45·0° F.; unheated area 43·7° F.; difference = 1·3° F.

Thus it was found that whether the heaters were lit or not, the thermometer at 3 inches in the heated area recorded a higher temperature than that outside it, showing that vegetation, particularly exposed grass, is able, under certain conditions, to influence air temperature to a considerable degree. The temperature differences obtained in this experiment were partly due to differences in height and density of the fruit trees and in the amount of grass beneath them. It follows that the vegetation factor must be taken into account in experiments on the effectiveness of orchard heating. This may be done by making comparisons between air temperatures with the heaters lit and not lit.

Another factor is the variation with time of the number of heat-units given out by the heaters. This may be estimated by ensuring that all heaters contain a uniform sample of fuel, sampling the fuel for analysis of calorific value, and keeping half-hourly records, when the heaters are lit, of the loss of weight of one or two representative heaters.

From the data obtained, variations in the heat output of all the heaters may be estimated, a knowledge of which is of value in accounting for differences in air temperatures inside and outside the heated area.

It may be useful here to state the conditions which, it appears, must be fulfilled in comparing fairly the temperatures of the air in a heated and an unheated plantation:—

(1) The thermometers inside and outside the heated area must be at the same height above ground level and in similar environments, especially with respect to exposed grass.

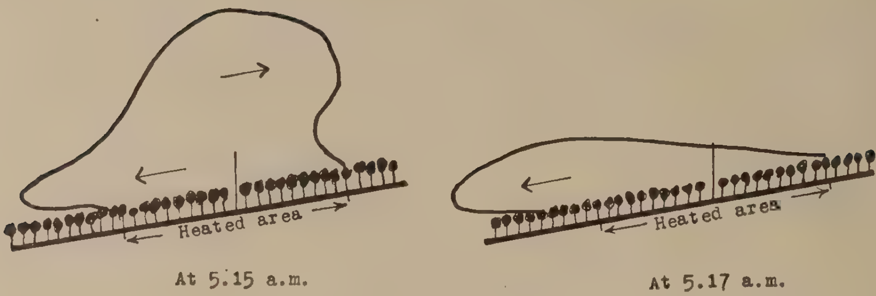

FIG. 18.

Showing the shape of the smoke cloud from orchard heaters at East Malling Research Station. The mast shown in the middle of the heated area is 50 feet high. The drawing is to scale but the slope is exaggerated. The shape of the cloud at 5.15 a.m. shows a katabatic wind blowing in the first 50 feet and the hill-top wind at 100 feet. Two minutes later the effect of the hill-top wind had disappeared and all the smoke began to move downhill in a katabatic wind.

(2) If there is any horizontal air movement, the thermometer outside the heated area must be on the windward side.

(3) The thermometer inside the heated area must be in the middle of four heaters, or at stated distances from them.

(4) The thermometers must be read simultaneously, or nearly so, and the times of doing so should be recorded. (Readings every 20 or 30 minutes are desirable, as the figures when plotted form fairly smooth curves which bear closer comparison than figures derived from isolated readings.)

(5) Both thermometers should if possible be at the same altitude above sea level, but environment is more important than altitude.

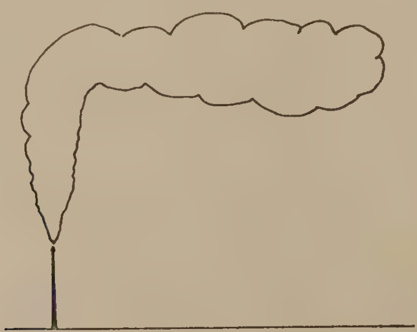

FIG. 19.

Smoke from a factory chimney in the Medway Valley on the evening of September 4th, 1937, indicating calm conditions to a height of 400 feet above which the south hill-top wind carried the smoke horizontally.

Observations on the smoke from orchard heaters, such as those illustrated in Fig. 18, showed that it was easily carried away by the most gentle air movement, such as a wind of 2 or 3 miles per hour, and this, of course, prevented any considerable rise in temperature of the orchard air. From this it is obvious that orchard heaters are likely to be most effective when the air in the orchard, and that above it to a height of several hundred feet, has no horizontal motion. The investigations described in the previous section showed at least some places where calm air is to be expected. The fact that the smoke from factory chimneys in the Medway valley, on certain radiation nights, e.g. that of September 4th-5th, 1937, has been observed to rise vertically to a height of several hundred feet, as shown in Fig. 19, shows that in large valleys the air may be calm. However, in the small valley at Leckford, Hampshire, the anemometers very rarely recorded a calm.

As stated by the writer in a previous paper (6) no conclusion could be drawn from these experiments as to the effectiveness of heaters in raising the orchard air temperature.

SUMMARY.

On nights when the air is not too humid, when the sky is clear and it is calm in the valleys, a light breeze of relatively warm air, called in this paper the hill-top wind, has been found on hill-tops. The breeze is able to influence the direction taken by a katabatic wind. The hill-top wind is even able, on the higher part of slopes facing it, to push the air uphill, and at a lower level to counterbalance or neutralize the force of the katabatic wind and thus produce a calm belt on the slope, called here the neutral zone. When the hill-top wind blows on a sloping field in a direction at right angles to the contour lines the katabatic wind moves underneath the hill-top wind in the opposite direction ; but when the hill-top wind blows obliquely on a sloping field the katabatic wind is deflected by it and the air in the neutral zone moves along the contours. The effect of these two winds on each other in certain other places is described.

The possibility of utilizing the hill-top wind for the avoidance of frost damage on relatively high land is suggested. A knowledge of the conditions affecting the direction of katabatic winds makes it possible to say where the air is likely to be sufficiently calm for orchard heating to be effective.

The differences in air temperature at a height of 3 feet above the ground, caused by differences in soil cover, that occur on some radiation nights in orchards and other fields have been investigated. The lowest minimum temperatures occur over grassland. The air above bare soil, and that in woods with a complete leaf canopy, is relatively warm, often 6° above that over grassland at a height of 3 feet above the ground.

Since vegetation influences air temperature it was necessary to eliminate this variable factor when studying the influence of altitude on air temperature. In this way it was shown that in undulating country the air temperature varies with altitude on most radiation nights, the highest places being the warmest. On some radiation nights, however, the reverse was found, showing that other factors, at present unknown, may influence air temperature.

It has been demonstrated that when a katabatic wind moves down a valley or a slope its temperature does not necessarily decrease, but is largely determined by the vegetation over which it passes.

It is suggested that in certain orchards containing long grass exposed to the sky in the blossoming period the danger of frost damage may be reduced by reducing the amount of exposed grass.

Experiments in micro-climatology and with orchard heaters showed that certain conditions must be fulfilled in making a just comparison between the air temperature in a heated plantation and that in an unheated one.

It should be emphasized that the investigation reported here is far from being complete. Until a great deal more becomes known no precise recommendations for the prevention of frost damage can be made.

ACKNOWLEDGMENTS.

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FIG. 4.

Photograph of a "river of mist" in a meadow in a valley at 7.18 p.m. on September 29th, 1937. The mist moves with the katabatic air and is shown moving downhill to the left over a 6′ stone wall.

FIG. 7.

Smoke cloud in the bottom of Leckford valley on a radiation night, showing katabatic wind 15 feet deep blowing to right, and north hill-top wind, above 15 feet, blowing to left.

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STUDIES ON POLLEN TUBE GROWTH IN PRUNUS*

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THE pollinations described in the present paper were carried out on trees grown under controlled conditions in a greenhouse. The flowers were emasculated, and when the stigmas were receptive, usually three or four days later, they were hand-pollinated.

Pollen tube growth in the cherry Noir de Schmidt was investigated. This variety was treated in three ways: (1) normal self-pollination, which served as a control; (2) self-pollination in which, before pollination, the styles were treated with the growth promoting substance phenylacetic acid, 1 per cent. being applied in lanolin; and (3) cross-pollination with the compatible variety Red Cluster.

The following pollinations in plums were investigated: (1) Coe's Golden Drop self-pollinated, (2) Coe's Golden Drop pollinated with Bryanston Gage, (3) Cambridge Gage pollinated with Late Orange and (4) Cambridge Gage pollinated with President.

The following inter-specific pollinations in plums were also investigated: (1) *Prunus divaricata* var. Myrobalan Yellow, a diploid, pollinated with *Prunus domestica* var. Victoria, a hexaploid, and (2) *Prunus domestica* var. Victoria pollinated with *Prunus divaricata* var. *Pissardii*, a diploid.

METHODS.

The pistils and styles were collected at different intervals after pollination, as described later, and were fixed in Flemming's and Karpechenko's fluids and in a mixture of equal parts of 95 per cent. alcohol, glacial acetic acid and lactic acid. The fixed material was washed and dehydrated as usual and imbedded in paraffin wax. Sections were cut 10μ to 18μ thick. The cherry pistils fixed in the alcohol:acetic acid:lactic acid mixture were stained by Haidenhain's iron alum haematoxylin method, using 0·5 per cent. and 0·25 per cent. haematoxylin solutions. The staining was quite satisfactory.

The plum styles and pistils fixed in Flemming's and Karpechenko's fluids were stained with safranin, occasionally using iodine as a mordant, and also with Delafield's haematoxylin and iron alum haematoxylin. Material fixed in Flemming's and stained with safranin gave the most satisfactory results. Delafield's haematoxylin gave good results with the plum Myrobalan Yellow

* Thesis in part accepted for the M.Sc. degree in Horticulture at the University of London.

where iron alum haematoxylin gave very poor results. The pistils of Myrobalan Yellow were fixed in Karpechenko's fluid only.

Cotton blue staining was tried with both cherry and plum styles, but the results were not satisfactory; moreover, the lactic acid in the stain makes manipulation of the preparation rather difficult.

Examination was made in serial paraffin sections of the pistils and styles. For measuring very short pollen tubes an ocular micrometer was used and longer ones were measured with the help of the mechanical stage.

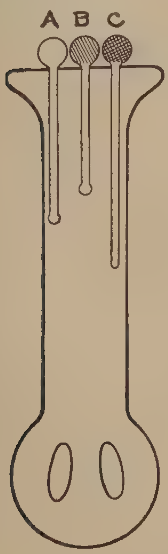
POLLINATIONS AND RESULTS.

I. CHERRIES.

Variety, Noir de Schmidt—(1) normal self-pollination; (2) self-pollination in which, before pollination, the styles were treated with phenylacetic acid; and (3) cross-pollination with the compatible variety Red Cluster. Pistils collected four days after pollination.

The detailed results of these pollinations are set out in Table I.

No significant differences were observed between (1) and (2). The proportion of pollen tubes which penetrated the styles and their rates of growth were approximately the same in the treated and in the control styles. Those tubes which penetrated the style became arrested in the stylar tissue sooner or later and most of them appeared to swell up at their ends, as shown in the photograph of a longitudinal section reproduced in Fig. 1, Plate I. In the compatible cross-pollination the growth rate was much more rapid than in (1) and (2), as can be seen from Table I and text Fig. 1. Very rarely one or two fruits were



TEXT FIG. 1.

Cherry Noir de Schmidt, 4 days after pollination.

A. The longest pollen tube (4·7 mm.) in normal self-pollination.

B. The longest pollen tube (3·8 mm.) in self-pollination when the styles were treated with phenylacetic acid.

C. The longest pollen tube (6·1 mm.) in the cross-compatible pollination with Red Cluster.

TABLE I.

Self- and Cross-pollination in Noir de Schmidt Cherry.

Material. Cherry.	No. of pistils examined.	Pollen tubes penetrating styles. No.	Pollen tubes penetrating styles. Lengths.	Average no. of pollen tubes per style.	Length of longest pollen tube. mm.	Average length of style. mm.	No. of flowers pollinated.	No. of fruits matured.	Per cent. fruits matured.
(1) Noir de Schmidt selfed (4 days after pollination).	3	34	·01—0·5 mm.	19·7	4·7	10·4	1,617	2	0
		2	·51—1·0 „						
		1	1·01—1·5 „						
		9	1·51—2·0 „						
		3	2·01—2·5 „						
		0	2·51—3·0 „						
		2	3·01—3·5 „						
		0	3·51—4·0 „						
		5	4·01—4·5 „						
		3	4·51—5·0 „						
		Total 59							
(2) Noir de Schmidt selfed and treated with phenylacetic acid (after 4 days).	3	34	·01—0·5 mm.	21·3	3·8	10·4	84	0	0
		4	·51—1·0 „						
		10	1·01—1·5 „						
		11	1·51—2·0 „						
		2	2·01—2·5 „						
		2	2·51—3·0 „						
		0	3·01—3·5 „						
		1	3·51—4·0 „						
		Total 64							
(3) Noir de Schmidt pollinated with Red Cluster (after 4 days).	3	51	·01—0·5 mm.	—	6·1	10·4	54	10	19
		4	·51—1·0 „						
		4	1·01—1·5 „						
		1	1·51—2·0 „						
		1	2·01—2·5 „						
		0	2·51—3·0 „						
		2	3·01—3·5 „						
		2	3·51—4·0 „						
		3	4·01—4·5 „						
		1	5·01—5·5 „						
		1	5·51—6·0 „						
		1	6·01—6·5 „						
		Total 71*							

* The full number of pollen tubes could not be counted owing to technical difficulties.

TABLE II.

Self- and Cross-pollination in Plum Varieties.

Material. Plum.	No. of pistils examined.	Pollen tubes penetrating styles. No.	Pollen tubes penetrating styles. Length.	Average no. of pollen tubes per style.	Longest pollen tube. mm.	Average length of style. mm.	No. of flowers pollinated.	No. of fruits matured.	Per cent. fruits matured.
(1) Coe's Golden Drop selfed (8 days after pollination).	1 (s)	3	·01—0·5 mm.	—	—	10·4	1,668	2	0
		0	·51—1·0 „						
		0	1·01—1·5 „						
		4	1·51—2·0 „						
		1	2·01—2·5 „						
		0	2·51—3·0 „						
		1	3·01—3·5 „						
		Total 9							
(2A) Coe's Golden Drop pollinated with Bryanston Gage (5 days).	1 (s)	7	·01—0·5 mm.	—	—	10·4	343	66	19
		0	·51—1·0 „						
		3	1·01—1·5 „						
		4	1·51—2·0 „						
		3	2·01—2·5 „						
		2	2·51—3·0 „						
		2	3·01—3·5 „						
		Total 21							
(2B) Cambridge Gage pollinated with Late Orange (9 days).	1 (s)	7	·01—0·5 mm.	—	—	10·5	733	16	2
		1	1·01—1·5 „						
		2	1·51—2·0 „						
		1	2·01—2·5 „						
		1	2·51—3·0 „						
		2	3·01—3·5 „						
		1	3·51—4·0 „						
		Total 15							
(2C) Cambridge Gage Pollinated with President (7 days).	2	71	·01—0·5 mm.	37·5	6·8	10·5	881	14	2
		0	1·01—1·5 „						
		1	2·01—2·5 „						
		1	3·01—3·5 „						
		1	3·51—4·0 „						
		1	6·51—7·0 „						
		Total 75							

(s) denotes upper half of the style only.

obtained after self-pollination, while after the compatible cross-pollination, 19 per cent. of fruits reached maturity, which is a good crop for this cherry.

The following additional growth promoting substances were also used on Noir de Schmidt: Naphtholacetic acid—66 pistils treated and self-pollinated; indolylacetic acid—25 pistils treated and self-pollinated; phenylacetic acid—42 pistils treated and not pollinated. No fruit set as a result of any of these treatments. A further 22 pistils of Noir de Schmidt were treated with phenylacetic acid and pollinated with Geant d'Hedelfingen; 4 fruits matured. From the same cross, without treatment with phenylacetic acid, 26 pistils being pollinated, 6 fruits set and matured.

II. Plums.

1. *Incompatible Pollinations.*

Coe's Golden Drop was self-pollinated and examined eight days after pollination. The results are given in Table II. All the pollen tubes seemed to become arrested in the stylar tissue. Such tubes often appeared to swell up at their ends as shown in Fig. 2, Plate I. One or two fruits have been very rarely obtained following this self-pollination (see Table II).

2. *Compatible and Partially Compatible Pollinations.*

(a) Coe's Golden Drop was pollinated with Bryanston Gage and examined five days after pollination. (b) Cambridge Gage was pollinated with Late Orange and examined nine days after pollination. (c) Cambridge Gage was pollinated with President and examined seven days after pollination. The results are given in Table II.

As shown in the Table, Coe's Golden Drop, when pollinated with Bryanston Gage, set a good crop of fruit, 19 per cent. of fruits reaching maturity, but Cambridge Gage set very few fruits when pollinated with Late Orange and President.*

In these compatible and partially compatible pollinations, in addition to normal pollen tubes which travel the full distance of the style and effect fertilization, tubes also occur which become arrested in growth in the stylar tissue and swell up at their ends, as shown in Fig. 2, Plate I for incompatible pollinations. Occasionally normal tubes and abnormal or incompatible tubes with swollen ends have been noticed side by side.

III. Inter-specific Pollinations.

1. *Prunus divaricata* var. Myrobalan Yellow, a diploid, was pollinated with *Prunus domestica* var. Victoria, a hexaploid, and examined seven days after pollination.

* In the reciprocal crosses, Late Orange pollinated with Cambridge Gage and President pollinated with Cambridge Gage, a full crop of fruit sets and matures (Crane and Lawrence (2)).

TABLE III.

Inter-specific Pollinations in Prunus *Species.*

Material. Prunus species.	No. of pistils examined.	Pollen tubes penetrating style. No.	Pollen tubes penetrating style. Length.	Average no. of pollen tubes per style.	Length of longest pollen tube. mm.	Length of style. mm.	No. of flowers pollinated.	No. of fruits matured.	Per cent. fruits matured.
(1) *Prunus divaricata* pollinated with *Prunus domestica* (7 days).	2	21 0 3 1 Total 25	·01—0·5 mm. ·51—1·0 „ 2·51—3·0 „ In ovary chamber	12·5	>5·6	5·6	179	11	6
(2) *Prunus domestica* pollinated with *Prunus divaricata* (3 days).	1	116 1 2 2 3 2 Total 126	·01—0·5 mm. 1·51—2·0 „ 2·01—2·5 „ 2·51—3·0 „ 3·51—4·0 „ 5·01—5·5 „	126	5·5	12·5	136	21	15

A pollen tube reaching the ovary chamber was noticed at this stage as shown in Fig. 3, Plate I. The number of fruits which matured in this inter-specific pollination amounted to 6 per cent.

2. *Prunus domestica* var. Victoria, a hexaploid, was pollinated with *Prunus divaricata* var. *Pissardii,* a diploid, and examined three days after pollination.

Pollen tubes penetrating about half the length of the style were observed at this stage. The length of the style in Victoria is more than double that of the style in Myrobalan Yellow. Hence the growth-rate of a diploid pollen tube in a hexaploid style is much greater than that of a hexaploid pollen tube in a diploid style, as could be seen from these reciprocal inter-specific pollinations. *Prunus domestica* var. Victoria pollinated with *Prunus divaricata* var. *Pissardii* set 15 per cent. fruits that reached maturity.

The results of both these inter-specific pollinations are summarized in Table III.

DISCUSSION.

Crane and Lawrence (2) and Crane and Brown (4), on the results of their extensive pollination and breeding experiments in cherries and plums, showed that the phenomenon of incompatibility in cherries can be expressed in terms of a series of multiple allelomorphs as S1, S2, S3, S4, etc., and that the problem of incompatibility is genetically simple in the diploid sweet cherries but more complex in the hexaploid plums. In sweet cherries, they have amply confirmed the oppositional factor hypothesis, i.e. that under normal conditions pollen cannot function in the style of a plant carrying the same factors as the pollen.

Afify (1) made some histological studies of pollen tube growth in cherries and plums and was able to distinguish between different pollen genotypes by the behaviour of the pollen on germination and the subsequent growth of the pollen tubes in the style. Thus, in the diploid sweet cherries he found three types of pollen grains, (a) those which failed to germinate, probably owing to generational sterility, (b) those which produced very short tubes which ultimately bent upwards and ceased to grow and (c) those which travelled the whole length of the style and effected fertilization. In hexaploid plums he found five types of pollen grains, (a) those which did not germinate, (b) those which developed short tubes which ultimately bent upwards and ceased to grow, (c) those with tubes which grew to a length of about a quarter of that of the style, (d) those with tubes which grew to a length of about half that of the style and (e) those with tubes which reached the ovary.

It will be seen from the present investigation that in addition to pollen grains which failed to germinate, grains that produced pollen tubes of very varied lengths penetrating the style occurred in all cases, whether the pollen

or the style belonged to a diploid or a hexaploid plant, or whether the pollination was a compatible or an incompatible one. In every case it is seen that some tubes were very short, some quite long, and that all degrees of intermediates occurred between them, whether the styles were examined at short or long intervals after pollination. In both cherries and plums, especially in compatible and partially compatible pollinations, at least two pollen genotypes could easily be distinguished, (a) those with tubes which were arrested in growth in the style and (b) those with tubes which travelled the whole length of the style.

SUMMARY.

1. Histological studies have shown that in the self-incompatible cherry Noir de Schmidt, treatment of the styles with phenylacetic acid has no accelerative effect on pollen tube growth or fruit development. The proportion and rate of growth of pollen tubes which penetrated the styles were found to be approximately the same in the treated and untreated styles following self-pollinations. Treatment of the styles with naphtholacetic and indolylacetic acids also had no effect on the setting or development of fruit.

2. In the self-incompatible plum, Coe's Golden Drop, when self-pollinated, the pollen tubes are arrested in the stylar tissue and their ends often swell up.

3. Plum styles pollinated with compatible and partially compatible varieties were also investigated. It was found that in addition to pollen tubes which travel the full length of the style and effect fertilization, tubes also occur which are arrested in the stylar tissue and swell up at their ends, thus indicating the existence of two pollen genotypes.

4. Inter-specific pollinations were also studied. In *Prunus divaricata* (diploid) pollinated with *Prunus domestica* (hexaploid), pollen tubes were observed in the ovarian cavity seven days after pollination. From this cross 6 per cent. of fruits set and matured. In the reciprocal cross 15 per cent. of fruits reached maturity.

5. The rate of growth of a diploid pollen tube in a hexaploid style is much more rapid than that of a hexaploid pollen tube in a diploid style.

ACKNOWLEDGMENTS.

The author is grateful to Mr. M. B. Crane under whose direction this work was carried out and to Sir Daniel Hall for the facilities provided for it at the John Innes Horticultural Institution.

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PLATE I.

FIG. 1.

Cherry Noir de Schmidt. Self-pollination in which, before pollination, the pistils were treated with phenylacetic acid. Length of the pollen tubes = 1·7 and 1·8 mm. respectively. Length of the style = 10 mm. × 235.

FIG. 2.

Plum Coe's Golden Drop, self-pollination. Length of pollen tube = 2 mm. Length of style = 10·4 mm. × 360.

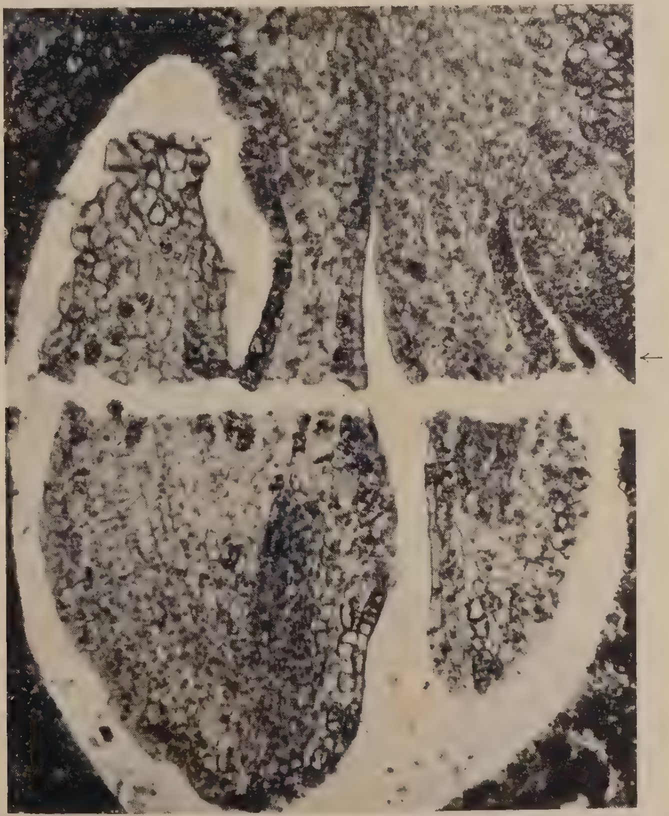

FIG. 3.

Inter-specific pollination. *Prunus divaricata* var. Myrobalan Yellow, a diploid, pollinated with *Prunus domestica* var. Victoria, a hexaploid. Pollen tube 7 days after pollination in the ovary chamber. × 287.

← = pollen tube.

THE EFFECT OF LEAD ARSENATE AND COPPER CARBONATE SPRAYS ON THE QUALITY OF ORANGES

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INTRODUCTION.

AMONG the insect pests that cause considerable loss of fruit to citrus growers in South Africa are the False Codling Moth, *Argyroploce leucotreta* Meyer, the Mediterranean Fruit-fly, *Ceratitis capitata* Wied., the Natal Fruit-fly, *Ceratitis* (*Pterandrus*) *rosa* Ksh., and the American Bollworm, *Heliothis armigera* Hubn. (*obsoleta* Fabr.). Both Fruit-flies deposit their eggs within the albedo of the citrus fruit by puncturing the rind. The False Codling Moth lays its eggs on the surface of the fruit and the young larvae eat their way in through the rind, while the Bollworm larvae eat mainly the outside as well as portions of the very young fruits.

Over a three-year period in the Eastern Transvaal, one of the main citrus-producing areas in the Union of South Africa, the second author found that in Washington Navel oranges in orchards where fairly thorough orchard sanitation was practised, the total percentage of almost full-sized fruit that dropped from the trees varied from 5 to 15 per cent., depending on climatic conditions and orchard location. In this dropped fruit the loss due to False Codling Moth was from 1 to 6 per cent. of the total crop, and that due to both species of Fruit-fly was from 0·5 to 2 per cent.

With Valencia oranges, which ripen some four months later than Navels and are not so subject to mature fruit drop, only 3 per cent. of the total crop dropped, 1 per cent. having False Codling Moth larvae on it, and the amount of fruit attacked by Fruit-fly was negligible. Ripley and Hepburn (13) reported that in portions of the Kat River Valley, owing to Fruit-fly, "losses up to 10 per cent. frequently occur . . . while much greater losses are occasionally experienced". In trapping experiments conducted by the second author with three types of bait in the Eastern Transvaal over a period of years, the Mediterranean Fruit-fly and the Natal Fruit-fly were found to occur in about equal numbers in the orchard; but no evidence is available to indicate whether each does the same amount of damage. The damage caused by Bollworm is exceedingly variable; in some years the infestation is negligible in all citrus areas in the Union, while at other times, such as in 1934 in the Lowveld, the crop in some orchards may be reduced by as much as 80 per cent. of the

normal, and even mature Valencia oranges, still hanging at the time the young fruit was pea-size, were severely attacked, the worms eating their way right through the fruit.

For many years growers of deciduous fruit have been using the Mally Fruit-fly bait, as recommended in 1915 for the control of the Mediterranean Fruit-fly, the bait being sprinkled on the foliage with a hand-syringe (spot-spray). It has also been known that if lead arsenate (the toxic material in the Mally bait) is cover-sprayed on citrus trees the fruit then becomes insipid. Ripley and Hepburn (12), seeking a non-arsenical poison for Fruit-fly, found that sodium fluosilicate was promising, its toxicity to the Natal Fruit-fly being sixteen times that of lead arsenate. They found no damage to citrus foliage in preliminary trials at concentrations of spray sufficiently high to insure adequate toxicity to the fly, but felt that further investigations were required to determine fully the affect of this material on the tree and on the flavour of the fruit.

During the eradication campaign against the Mediterranean Fruit-fly in Florida, in 1929, complete cover-sprays of lead arsenate were repeatedly applied to citrus trees. The disappearance of the acid from the fruit, owing to the arsenic, made this compound such an unsatisfactory one to use on citrus trees that other insecticides were investigated by Miller and McBride (10), emphasis being placed on copper salts. The toxicity of copper carbonate was found to equal that of lead arsenate for the Mediterranean Fruit-fly. Ripley and Hepburn (12) found that copper carbonate was not sufficiently toxic to the Natal Fruit-fly, however, to be useful as a poison for the control of this insect.

The control of False Codling Moth in citrus presents a problem to entomologists. Orchard sanitation is the only control measure recommended at present and, in general, gives a satisfactory degree of control. However, particularly where this is neglected, it is possible that in instances of severe infestation the use of an insecticidal cover-spray may be beneficial.

The present paper reports the results of investigations with lead arsenate and copper carbonate sprays during the period 1932-36 in the Eastern Transvaal. The results obtained with fluorine compounds were such as to warrant further comprehensive experiments, which are now in progress elsewhere in the Union, and they will no doubt be reported in later papers.

PREVIOUS WORK ON EFFECT ON TREE AND FRUIT.

Many workers have shown that the application of arsenic compounds to citrus trees results in a reduction of the acid content of the fruit juice, one of the first reports of early maturing of fruit from trees sprayed with arsenic compounds having appeared in 1894. Until 1921 not much scientific literature on this subject had appeared, Gray and Ryan (5) having been among the first to present the results of investigations.

An excellent review of the literature pertaining to the effect of lead arsenate on the soluble solids and acid content of the juice of citrus fruits from trees to which this insecticide had been applied was given by Miller, Bassett and Yothers in 1933 (11). These workers found that 30 lb. of lead arsenate applied as a spray to the soil under a grape-fruit tree in sixty doses produced no noticeable effect on the tree, fruit or undergrowth. An orange tree receiving 7 lb. of lead arsenate in a similar manner also showed no effect, even though a great excess of arsenic trioxide was found in the soil to a depth of fourteen inches after 115 inches of rain had fallen during the following eighteen months. Thus, the danger of "soil poisoning" for citrus through the continuous use of arsenical sprays was shown to be negligible. Jones and Hatch (7) have shown that orchard welfare is not directly affected by the inevitable accumulations of inorganic spray materials in the soil, as these penetrate to a depth of only six to eight inches; and while cover-crops might fail, danger to human beings or other animals arises only after the trees have been removed and attempts are made to grow annual crops. A year after trees had been sprayed by Miller, Bassett and Yothers (11) a diminishing amount of water-soluble and insoluble arsenic was found to be still present on the leaf surfaces, and varying amounts of arsenic (from a trace upwards) were found in blossoms, small fruits and in the wood of the tree. The spray was applied to one half of the tree, the other half being used as a control. It was shown that contrary to the views of Gray and Ryan (5), Juritz (8) and Copeman (2) the effect of the arsenic is not systemic, thus confirming the previous contention of Yothers (15), and also the work of Miller and McBride (10), who found that the effect of arsenic is carried through the leaves and twigs to the immediately neighbouring fruit. When only one individual fruit was treated, that fruit alone became affected. Miller, Bassett and Yothers also found that very small quantities of spray stimulated leaf respiration and catalase activity and reduced the acid in the juice of the fruit. Their further findings that lead arsenate, when applied in small quantities, increased, and in larger amounts, decreased the soluble solids in the juice, will be discussed later. When only 0·01 gm. of As_2O_3 per 10 gm. of leaves was present, the soluble solids/acid ratio of the juice was a little above normal, but larger quantities of the arsenate resulted in an insipid juice. The effect was shown immediately, persisted to a slight degree for eighteen months, and had entirely disappeared after two and a half years, depending on the presence of soluble arsenic on the actively growing leaves of the tree.

The only reference found by the present authors as to the effect of copper or copper compounds on the citrus tree and its fruit is that of Miller and McBride (10), who point out that, while no extensive data were available as to the effect of copper carbonate on the tree and fruit quality, Bordeaux sprays had been used

for years for curing "dieback", that copper salts had been applied to citrus groves as "soil conditioners", and that in all this no effect on the fruit quality had been noticed. Bordeaux and other copper-containing sprays have also been used extensively for the control of fruit rots of oranges still on the tree.

LAY-OUT OF THE EXPERIMENT.

Through the courtesy of Mr. P. Greathead, of Barclay Vale, in the Eastern Transvaal, a large block of Washington Navel orange trees on rough lemon stock, fifteen years old at the time, was placed at the disposal of the present authors for any work they wished to carry out. The co-operation of Mr. Greathead in these experiments is acknowledged with great appreciation.

Much attention has lately been focussed on the lay-out of experiments with fruit trees, and there is no doubt that the evaluation of data obtained from many experiments in the past is open to the severest criticism owing to what, in the light of present-day knowledge, was a faulty or "biased" lay-out of the plots receiving differential treatments. To render statistical treatment of the data obtained possible, it is essential that experiments should be laid out in accordance with the accepted principles that apply to horticultural experimentation (see Hoblyn (6)). Even where the results obtained seem to show differences large enough to leave no doubt in the worker's mind as to their significance, without statistical treatment, caution must be used in accepting them unless similar results are obtained repeatedly over a period of years in different localities approximating to the first one in tree environmental conditions.

With reference to previous work on the effect of sprays on citrus fruit quality, it is contended here that too much importance should not be attached to the actual data or figures presented. This is on account of the failure of most workers to consider their plot lay-out with a view to a statistical interpretation of the results. The trends or indications of their findings, however, are of great value. The present authors themselves plead guilty to having failed to give the fullest consideration to the lay-out of the experiment to be described here, and in the presentation of the data special attention will be given in interpreting them to any differences that may have been due to chance and others that were the results of the differential treatments applied.

The accompanying diagram (Fig. 1) shows the blocks and plots used in the experiment. Owing to seepage from the constantly filled main irrigation furrow, the trees in the upper part of the orchard were rather variable; most of the tree spaces marked as blanks were filled with trees in poor condition in 1932, such trees having been removed subsequently. Each of the three blocks consisted of three plots with a single row of trees as a control between each plot. The plots consisted of two rows of twelve tree-spaces each. The three original treatments were repeated once in each block for the first year; in the second,

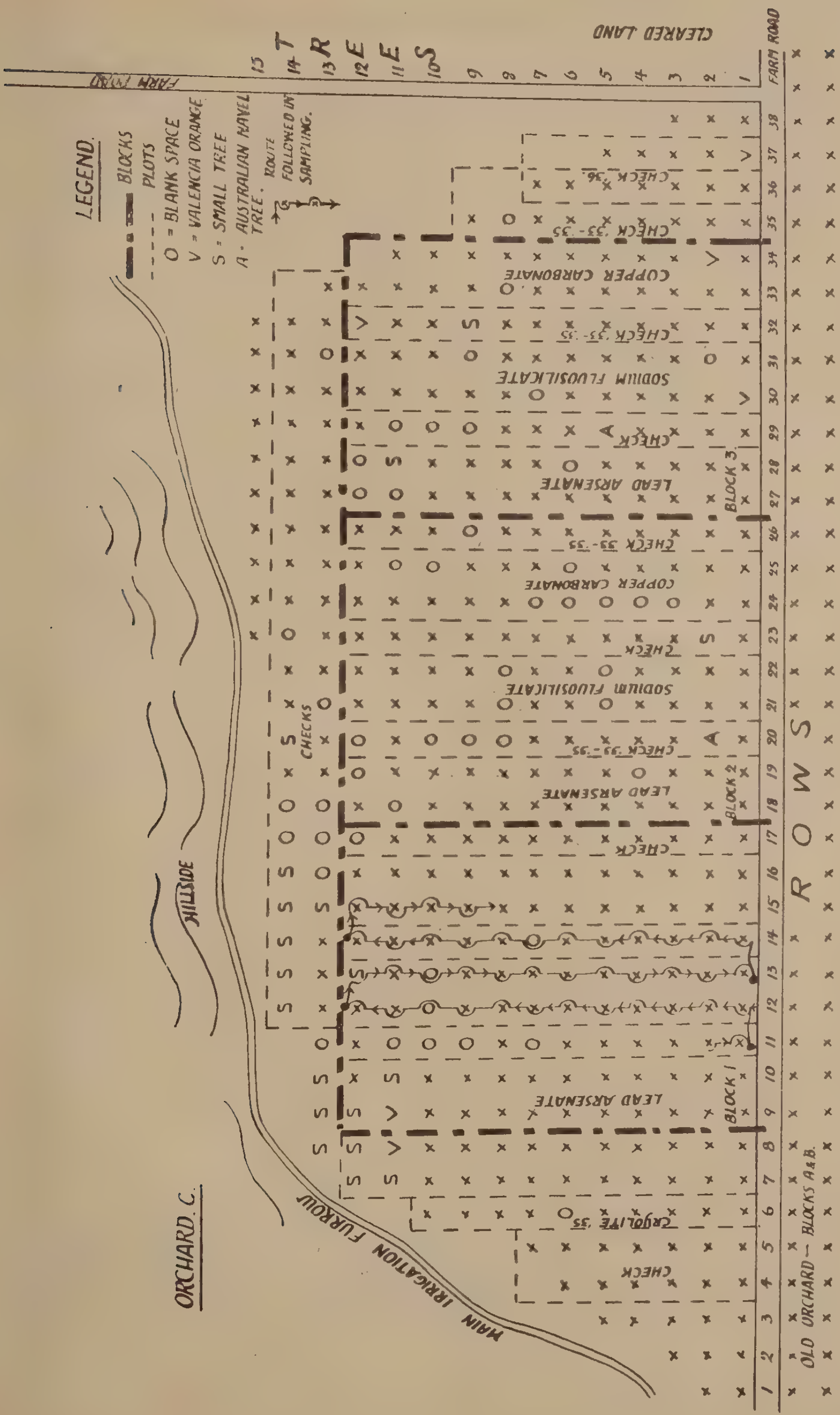

FIG. 1.
Lay-out of the blocks and plots in the orchard

only the trees in blocks 2 and 3 were sprayed, those in block 1 being used to observe the residual effect of the first-year treatments. In the third year only the trees in the plots in block 3 were sprayed, those in blocks 1 and 2 serving to indicate the residual effect of spraying after two and one years respectively. In the fourth year certain modifications were made in the use of the plots, a fourth treatment having been introduced, for which certain control rows and other previously-treated rows in block 1 were used.

SPRAYS AND SPRAYING SCHEDULES.

Copper carbonate, sodium fluosilicate, lead arsenate, and, in the last year, natural cryolite (sodium fluoaluminate) were the insecticidal compounds tested for their influence on fruit quality.

Since the start of the experiments the question of baiting for Fruit-fly was uppermost in the mind of the authors, the first year's treatment consisted of "spot-sprays", 21 fluid ounces of liquid being applied per tree with a hand-syringe at each application. This baiting was started on February 2nd, when the fruit was almost full-sized but hard, dark green and still extremely immature internally. It was continued at fortnightly intervals, except when rain delayed it for a day or two, till June 21st, when the fruit had been fully coloured for some time and was almost over-mature internally. Eleven spot-spray applications had been made by that time.

The formulae used and other details of treatment were as follows:—

1933 Formula (based on the Mally bait):—

Toxic compound	2 oz.	
Treacle (cane)	½ gal.	(Imperial.)
Water	4 gal.	(,,)

Thus, to each tree the following respective approximate amounts of toxic compounds were applied:—

After 3 spot-sprays ..	¼ oz.	(Fruit analysed.)
After 7 spot-sprays ..	½ oz.	(,, ,,)
After 11 spot-sprays ..	1 oz.	(,, ,,)

1934 Formula:—

Toxic compound	1 lb.	
Calcium caseinate (spreader) ..	1 lb.	
Water	100 gal.	(Imperial.)

The insecticidal compounds were applied as cover-sprays, approximately four gallons per tree per application being used. This change was made to obtain a greater effect of the toxic compounds on the tree and the fruit; for not

only was it seen from the fruit analyses made after the previous season's spot-spraying that even such a substance as lead arsenate (known to have a decided influence on the acid content of the fruit juice in the small quatities applied in spot-spraying) did not greatly influence the fruit quality, but also that the all-important possibility of controlling False Codling Moth and Bollworm by means of cover-sprays required consideration.

Because Ripley and Hepburn (12) had shown that sodium fluosilicate could be used at much lower concentrations than lead arsenate to produce the same toxic effect with Natal Fruit-fly, a compromise was made between the Mally bait and their findings in the amount of toxic compound in the cover-sprays. Between February 27th and May 23rd each tree received the following approximate amounts of toxic compound:—

After 2 cover-sprays ..	1¼ oz.	(Fruit analysed 1 week later.)
After 3 cover-sprays ..	2 oz.	(,, ,, 2 weeks ,,)
After 4 cover-sprays ..	2½ oz.	(,, ,, 5 ,, ,,)

1935 FORMULA:—

Toxic compound	2 lb.	
Calcium caseinate	1 lb.	
Water	100 gal.	(Imperial.)

The amount of toxic compound in the cover-sprays was doubled; for, apart from the expected reduction in acid content of the fruit sprayed with lead arsenate, no great differences could as yet be noticed in the fruits from trees sprayed with other materials.

Four cover-sprays were applied between February 4th and April 30th, each tree receiving the following approximate amounts of toxic compound:—

After 3 cover-sprays ..	3¾ oz.	(Fruit analysed 1 week later.)
After 4 cover-sprays ..	5 oz.	(,, ,, 5 weeks ,,)

Figures for the total quantity of toxic compound applied per tree over the four-year period can be obtained from the Tables given. It should be noted that a cumulative influence of these amounts cannot be presumed, as, apart from the washing off of the spray by rain, its removal by the normal shedding of leaves is considerable but cannot be estimated.

FRUIT ANALYSES.

Since de Villiers (3) had shown that in uniformly treated orchards the Brix No./acid ratio of the juice of oranges varies according to the side of the tree from which the fruit is picked, and that the position of the fruit on the tree, while hardly influencing the ratio, markedly influences the total acid and total

soluble solids in the juice, care was taken that all samples analysed were representative of the total fruit in the respective plots.

In order to obtain some idea of the variation which would be found in analysing the samples taken in the orchard, analyses were made in 1932 before any spraying had been done. Eleven plots of five adjacently grouped trees each were selected at random over the whole orchard. The sample to be analysed from each plot consisted of twenty fruits of uniform size and colour, there being four from each tree, picked at approximately the same height from the outside of the tree, one fruit being taken from each of the north, south, east and west sides. The results of these analyses were:—

TABLE I.

Analyses of fruits.

	Sol. Solids.	Acid.	Ratio of Solids to Acids.
Mean of 11 samples ..	12·5% ± ·09	0·99% ± ·08	12·7 ± 0·17
Coefficient of variability ..	4	35·6	7·5
Highest determination ..	13·1%	1·15%	14·8
Lowest determination ..	11·7%	0·84%	10·5

Allwright (1) considers that the minimum number of orange fruits per sample should be fifteen. He found that by increasing the size of the sample from sixteen to thirty-two fruits, the coefficient of variability for juice percentage dropped from 4·5 to only 4 per cent., and that of the soluble solids/acid ratio diminished from 7 to 6 per cent. Although de Villiers (3) found that ten-orange samples yielded analysis figures approximating very closely to the "true" or "ideal" values, his graphs show that it was only after the samples contained twenty-five oranges that the soluble solids, acid, and the ratio values gave straight lines when plotted. Since it was merely a question of extra labour involved, it was decided that samples of twenty-four oranges each should be used in the analysis recorded here.

One sample was picked from each twelve-tree row; about thirty or more fruits, of approximately equal size, of the same degree of colour, and from about the same height on the tree, were taken while walking from tree to tree in a zig-zag manner, as shown in the diagram (Fig. 1). Each sample as picked was afterwards spread on a table, and twenty-four of the most uniform fruits as regards size and colour were selected for analysis. The soluble solids content of the juice was determined with a Brix hydrometer, and the acid content by titration with sodium hydroxide solution.

Three analyses at approximately five-weekly intervals were made during each of the four years: (1) when the fruit was full-sized but still quite green;

(2) after the fruit had started to colour; and (3) when all the fruit had been well coloured for some time. In 1935, analysis No. 2 was omitted as the trees bore an extremely light crop following a severe infestation of Bollworm in 1934 which destroyed the small fruit just set.

RESULTS OBTAINED.

Although testing the effect of the four insecticides on fruit quality constituted one large experiment, the results for each compound will now be presented separately. Where six or more samples per treatment were available, the Probable Errors of the Averages and of the Differences are given to indicate the degree of certainty on which the interpretations of the results have been based, it being considered that where the Difference is three times greater than the P.E. of the Difference, such differences were due to the treatment applied and not to chance. The Probable Errors given in the preceding Table are small, as are those for the control rows over the four-year period. This indicates that the method of sampling was satisfactory. Since this was always done in the same manner it has been considered permissible to draw conclusions from certain differences, even though there were only two replications per treatment, when such differences appeared in more than one year and in the several analyses made each year. The accompanying Tables give the averages of some 350 individual samples analysed for fruit quality.

Lead Arsenate.

In Table II the averages of the analyses for soluble solids and acid in the fruit juice are given together with the ratio of these as determined for trees sprayed with lead arsenate.

It will be seen that the small amount of 1 oz. of lead arsenate per large tree, applied in eleven spot-sprays during the first year, started to show its effect after seven applications had been made, and at the end of the season the decrease of from 0·73 to 0·62 per cent. of acid caused an increase in the ratio from 15·3 : 1 to 17·8 : 1.

However, the residual effect of the 1933 spray on the leaves and branches of the trees (absent from the new crop set after the first year's spraying had been completed) was striking. Even two months before the normal crop was ready for harvesting the acid in the previously sprayed trees was 0·93 as compared with 1·19 per cent. in the controls; and this difference increased later in the season to 0·67 as compared with 0·97 per cent., while at the end of the season, the figures were 0·48 and 0·83 per cent. respectively.

The residual effect was still noticeable in the second year after spraying, but it was diminishing markedly. The rainfall for the year was thirty inches. At

TABLE II.

Effect of lead arsenate on fruit quality.

Treatment.	No. of Samples.	First Analysis.						Second Analysis.						Third Analysis.					
		% Sol. Solids.	*P.E.*	% Acid.	*P.E.*	Ratio of Solids to Acid.	*P.E.*	% Sol. Solids.	*P.E.*	% Acid.	*P.E.*	Ratio of Solids to Acid.	*P.E.*	% Sol. Solids.	*P.E.*	% Acid.	*P.E.*	Ratio of Solids to Acid.	*P.E.*
1933				March 8th.						May 10th.						June 28th.			
Control	11	10·1	·11	1·48	·03	6·8	·13	9·9	·19	0·83	·03	12·0	·16	11·1	·05	0·73	·04	15·3	·24
Spot Sprayed	6	10·1	·05	1·52	·03	6·6	·03	10·0*		0·75		13·4		11·0	·14	0·62	·02	17·8	·37
Difference		0	·12	**+0·04**	·04	**—0·2**	·13	**+0·1**		**—0·08**		**+1·4**		**—0·1**	·15	**—0·11**	·04	**+2·5**	·44
1934				April 6th.						May 10th.						June 28th.			
Control	11	10·0	·09	1·19	·02	8·5	·21	10·8	·04	0·97	·02	11·2	·27	12·0	·09	0·83	·02	14·5	·34
Cover Sprayed	4	10·4		0·98		10·7		11·1		0·68		16·3		12·0		0·53		22·6	
Difference		**+0·4**		**—0·21**		**+2·2**		**+0·3**		**—0·29**		**+5·1**		0		**—0·30**		**+8·1**	
Residual Effect from 1933	2	10·3		0·93		11·2		10·6		0·67		15·5		12·0		0·48		24·9	
Difference		**+0·3**		**—0·26**		**+2·7**		**—0·2**		**—0·3**		**+4·3**		0		**—0·35**		**+10·4**	
1935				April 8th.												June 10th.			
Control	11	10·1	·14	1·41	·03	7·2	·16							11·7	·12	1·10	·01	10·7	·24
Residual Effect from 1933	2	10·2		1·25		8·1								11·4		0·88		13·0	
Difference		**+0·1**		**—0·16**		**+0·9**								**—0·3**		**+0·22**		**+ 2·3**	
Residual Effect from 1934	2	10·1		0·96		10·1								11·9		0·53		22·4	
Difference		0		**—0·45**		**+2·9**								**+0·2**		**—0·57**		**+11·7**	
Cover Sprayed (2 years) ..	12	10·7		0·87		12·3								11·4		0·52		21·9	
Difference		**+0·6**		**—0·54**		**+5·1**								**—0·3**		**—0·58**		**+11·2**	
1936				April 17th.						May 8th.						June 26th.			
Control	8	9·6	·12	1·13	·01	8·6	·13	10·0	·03	0·87	·02	11·6	·20	11·6	·10	0·77	·02	15·2	·18
Residual Effect from 1933	2	9·6		1·08		8·9		9·9		0·88		11·3		11·6		0·76		15·4	
Difference		0		**—0·05**		**+0·3**		**—0·1**		**+0·01**		**—0·3**		0		**—0·01**		**+ 0·2**	
Residual Effect from 1934	2	9·8		1·10		8·9		10·2		0·89		11·4		11·9		0·72		16·5	
Difference		**+0·2**		**—0·03**		**+0·3**		**+0·2**		**+0·02**		**—0·2**		**+0·3**		**—0·05**		**+0·7**	
Residual Effect from 1935	2	9·3		0·62		15·0		9·8		0·46		21·1		11·2		0·34		32·7	
Difference		**—0·3**		**—0·51**		**+6·4**		**—0·2**		**—0·43**		**+9·5**		**—0·4**		**—0·43**		**+17·5**	

* Data for 1934 for this analysis obtained from three composite samples from 2 rows each, hence no probable errors calculated.

TABLE III.

Effect of copper carbonate on fruit quality.

Treatment.	No. of Samples.	First Analysis.						Second Analysis.						Third Analysis.					
		% Sol. Solids.	*P.E.*	% Acid.	*P.E.*	Ratio of Solids to Acid.	*P.E.*	% Sol. Solids.	*P.E.*	% Acid.	*P.E.*	Ratio of Solids to Acid.	*P.E.*	% Sol. Solids.	*P.E.*	% Acid.	*P.E.*	Ratio of Solids to Acid.	*P.E.*
1933		March 8th.						May 10th.						June 28th.					
Control	11	10·1	*·11*	1·48	*·03*	6·8	*·13*	9·9	*·19*	0·83	*·03*	12·0	*·16*	11·1	*·05*	0·73	*·04*	15·3	*·24*
Spot-Sprayed (syringe) ..	6	10·2	*·22*	1·52	*·01*	6·7	*·02*	10·2		0·83		12·3		11·3	*·06*	0·72	*·02*	15·6	*·21*
Difference		**+0·1**	*·25*	**+0·04**	*·03*	**—0·1**	*·13*	**+0·3**		**0**		**+0·3**		**+0·2**	*·08*	**—0·1**	*·05*	**+0·3**	*·32*
1934		March 8th.						May 10th.						June 28th.					
Control	11	10·0	*·09*	1·19	*·02*	8·5	*·21*	10·8	*·04*	0·97	*·02*	11·2	*·27*	12·0	*·09*	0·83	*·02*	14·5	*·34*
Residual Effect from 1933	2	10·1		1·28		7·8		11·1		1·10		10·1		11·7		0·90		13·0	
Difference		**+0·1**		**+0·09**		**—0·7**		**+0·3**		**+0·13**		**—1·1**		**—0·3**		**—0·07**		**—1·5**	
Cover Sprayed	4	10·2		1·15		8·8		11·1		1·00		11·1		12·0		0·91		13·2	
Difference		**+0·2**		**—0·04**		**+0·3**		**+0·3**		**+0·03**		**—0·1**		**0**		**+0·08**		**—1·3**	
1935		April 8th.												June 10th.					
Control	11	10·1	*·14*	1·41	*·03*	7·2	*·16*							11·7	*·12*	1·10	*·01*	10·7	*·24*
Residual Effect from 1933	2	9·9		1·50		6·6								11·4		1·11		10·2	
Residual Effect from 1934	2	10·0		1·42		7·0								11·3		1·08		10·5	
Cover Sprayed—2 years ..	2	10·0		1·31		7·6								11·2		1·01		11·1	
1936		April 17th.						May 8th.						June 26th.					
Control	8	9·6	*·12*	1·13	*·01*	8·6	*·13*	10·0	*·03*	0·87	*·02*	11·6	*·20*	11·6	*·10*	0·77	*·02*	15·2	*·18*
Residual Effect from 1934	2	9·5		0·98		9·7		10·0		0·81		12·3		11·1		0·72		15·4	
Residual Effect from 1935	2	9·0		1·01		8·9		10·0		0·85		11·8		10·9		0·68		16·1	

the end of the season, previously sprayed trees had 0·88 per cent. acid and a ratio of 13 : 1, as compared with 0·1 per cent. acid and a ratio of 10·7 : 1 for the controls. No residual effect on spot-sprayed trees was noticed in the third year after spraying.

When complete cover-sprays (2½ oz. of lead arsenate per tree) were applied to rows which had been spot-sprayed in the previous year, the effect on the acid became evident earlier in the season. By mid-season the acid in the fruit from sprayed trees had dropped from 0·97 to 0·68 per cent., and by the end of the season the sprayed fruits had a ratio of 22·6 : 1 as compared with 14·5 : 1 for the controls.

The first year residual effect of the cover-sprays was striking, reductions in acid from 1·41 to 0·96 per cent. and from 1·1 to 0·53 per cent. being found in early and late season tests respectively. A diminution in the effect was noticed in the second year after spraying. The second year cover-spraying caused a reduction from 1·41 to 0·87 per cent. early in the season, and from 1·1 to 0·52 per cent. late in the season.

The most noticeable residual effect was shown in 1936, by trees which had received one spot-spray and two cover-sprays during the previous three years. Early in the season their fruit had only 0·67 per cent. of acid as compared with 1·13 per cent. in the controls, and at the end of the season the acid had been reduced to 0·34 per cent., giving a ratio of 32·7 : 1, as compared with 0·77 per cent. acid and a ratio of 15·1 : 1 for the controls.

In the analyses made over the four years there was no indication that the lead arsenate spray had had any influence on the total soluble solids in the juice of the fruit ; the differences found were small and variable, being indiscriminately minus and plus.

Copper Carbonate.

Table III is included to show that in these experiments copper carbonate had absolutely no effect on the quality of the fruit from trees sprayed with it, the acid and total soluble solids in the fruit juice not having been altered through spraying. It was observed, however, that the fruit on trees cover-sprayed with copper carbonate, at 5 oz. per large tree, coloured up slightly earlier than fruit on unsprayed trees. This earlier colouring was not reflected in the analyses made.

FURTHER CHEMICAL ANALYSES OF THE FRUIT.

Through the courtesy of the Chief of the Division of Chemical Services certain representative samples of fruit from sprayed rows and from controls were analysed for their lead, arsenic and copper content. The main reason for

these analyses was to obtain some indication as to whether sprayed fruit contained toxic elements in excess of the minimum specified in the pure food and health laws of the Union and of other countries in which citrus fruit is consumed.

Arsenic. Paragraph No. 3 (2) of the Regulations published under Government Notice No. 572 of March 28th, 1930, of the Union Act 13 of 1929, provides that "No article of food shall contain, per pound or per pint, more than . . . . , or a hundredth (1/100) of a grain of arsenic (calculated as arsenious oxide, As_2O_3). . . ."

Analyses made in 1935 showed that the total As_2O_3 in the juice of fruit from trees sprayed for three years with lead arsenate, including 1935, was only 1/1,200 of a grain per pound, and only one-third of this very small quantity was found to be present normally in fruit from trees which had never been sprayed. Thus, even though arsenic enters the fruit from spray applied to the leaves of the tree, the quantity is so small as to present absolutely no danger to the health of persons consuming fruit from sprayed trees, provided they do not consume the rind. The amount of arsenic which would be present on the rind is very variable, depending on the adhesive power of the spreader used in the spray, on the rainfall between the last application and the time of harvesting, and on the treatment the fruit received before it was packed, such as washing, brushing and polishing.

Copper and Lead. Both in 1935 and 1936 analyses were made for copper and lead in the rind and juice of fruit from trees sprayed with copper carbonate and lead arsenate. The results showed that only indeterminable amounts (less than 1 p.p.m.) of these heavy metals were present; thus oranges from trees heavily sprayed with materials containing these metals can be regarded as quite safe for human consumption.

DISCUSSION.

Quality of fruit in this work has been measured by the total soluble solids and acid content of the juice of the fruit. Thus, when the percentage of either of these components changes after the application of the toxic compounds to the tree, it is said that the quality of the fruit has become affected. Whether the small changes recorded in some instances can be noticed in the actual taste of the fruit does not enter into this discussion; where the acid content is considerably reduced, the insipidity of the juice can easily be noticed, and if the fruit is low in soluble solids then the juice lacks body.

The practical aspect of the effect of insecticides on fruit quality is of paramount importance. Practically all citrus-producing countries have laws and regulations determining the standard of quality of the fruit that may be exported or offered for sale on its local markets. The grower's concern is that his fruit

shall comply with such standards at a time when the prices realized for the fruit are most favourable. In Florida, legislation was passed many years ago to prevent growers from spraying their citrus trees with arsenical sprays to reduce the acid in the fruit, thereby enabling them to place their fruit early on an empty market, and thus marketing fruit which, although complying with the regulations, was of poor eating quality.

When growers have to use insecticides and fungicides for the control of pests and diseases, and if such materials affect the fruit quality adversely, a change must be made in the materials or methods used, or in both. The degree of the effect is important in that the quality of the fruit from the same variety varies greatly under different climatic and environmental conditions. Certain varieties would be much improved in fruit quality if, for instance, either the soluble solids could be increased or the acid decreased, without having to leave the fruit on the tree until it becomes "over-mature", when the rind would be in such a condition as to make successful transport to distant markets difficult.

The use of arsenical materials as cover-sprays on orange trees should be avoided on account of the marked reduction they bring about in the acid content of the fruit juice, as already proved by many other workers and confirmed in the present experiment. The effect is not truly non-systemic, in that if the material is applied to leaves it will travel to neighbouring fruit ; but if applied to one side of a tree only, it will not affect the other side. Fruit that sets months after the spray has been applied will be markedly affected by the spray deposit still present on the leaves of the tree. This influence may last for two or three years, the length of its persistence depending on the amount of the toxic agent originally applied, on the rainfall, and on the time elapsing before the leaves on the tree at the time of spraying fall off.

Lead arsenate, applied as a spray in these experiments, had no influence at all on the total soluble solids in the juice. Miller *et al.* (11) claim that this is not so ; but if a line be drawn from point to point on their semi-logarithmic graph it will appear to be doubtful if, even on their own data, their conclusion is justified, particularly as their lowest soluble solids point was obtained when 30 lb. of lead arsenate was applied to one tree only. Nor can reliance be placed on the figures of Juritz (8), which show a reduction in reducing sugars ; for his method of sampling could not have given even approximately truly representative samples. Gray and Ryan (5), Copeman (2), and Takahashi (14) found no influence on the sugars in the juice subsequent to applications of lead arsenate. It is therefore claimed that the evidence is overwhelmingly in favour of the view that lead arsenate does not affect the soluble solids or sugars in the juice of oranges.

On occasion, growers in the Eastern Transvaal have sprayed their Valencia orange trees with lead arsenate to bring about a reduction in the acid content of the

fruit, in order to enable them to pick earlier than usual and so take advantage of favourable market prices. Where small quantities of spray were applied with hand-syringes the desired result was brought about, but much insipid and poor-eating quality fruit has resulted from too heavy and continued applications to the trees. In addition, growers must be warned that the tolerance for arsenic is calculated on the *whole* fruit, even though the rind of oranges is usually discarded ; and unless sprayed fruit is washed, growers offering such fruit for sale are liable to find themselves in conflict with the Health Authorities, both in South Africa and the United Kingdom.

The question naturally arises as to whether the two insecticides used in the experiment here reported would affect the fruit quality in a similar manner if they were applied as dusts. When substituting a dust for a liquid spray, about four times the quantity of the insecticidal agent contained in the spray is generally applied in the dust per tree. There is no reason to believe that the total effect would be any different, but the degree would probably be less. In curing Mottle-leaf of citrus, zinc oxide and zinc sulphate exert an influence in varying degrees, whether applied as dusts or sprays, the zinc being taken into the tree in solution, the moisture necessary for this coming from rains, dew, and the aqueous vapours released by the transpiring cells in and around the applied material on the leaf surfaces. Further work requires to be done to determine experimentally the effect of insecticidal dusts on the quality of citrus fruits.

SUMMARY.

The False Codling Moth, the Mediterranean Fruit-fly, the Natal Fruit-fly, and the American Bollworm are pests of oranges in South Africa.

Among the insecticidal agents experimented with and used for pest control are lead arsenate, copper carbonate, sodium fluosilicate and sodium fluoaluminate (cryolite). This paper reports the results obtained over a four-year period of the effect of lead arsenate and copper carbonate, applied as sprays to the tree, on the quality of Washington Navel orange fruits, as determined by the total soluble solids and acid content of the juice.

Details of the lay-out of the experiment, the sprays used, their schedule of application and the method of obtaining representative fruit samples are given in full.

Lead arsenate, applied as a spot-spray, at the rate of 1 ounce of lead arsenate per large tree, was responsible for a slight increase in the soluble solids/acid ratio in the juice of the fruit ; and when cover-sprays carrying 5 ounces of it per tree were applied, it caused a marked reduction in acid.

A residual effect of lead arsenate on acid reduction was evident in the two crops following the application of both spot- and cover-sprays. The effect of

lead arsenate on acid reduction was more marked on the crop of the year following the application of the spray than on the crop maturing at the time of application.

No evidence was obtained of any effect of lead arsenate on the total soluble solids in the fruit juice.

Copper carbonate, applied in five cover-sprays at the total rate of 5 ounces per large tree, showed no influence on either the total soluble solids or the acid content of the juice. A slight earliness in colouring of the fruit sprayed with this material was observed.

Chemical analyses showed that the quantity of arsenic present in the fruit juice following cover-sprays with lead arsenate was negligible.

No increase in the lead or copper content of the fruit juice above that normally found resulted from cover-sprays of lead arsenate and copper carbonate.

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THE SEASONAL CYCLES OF ASH, CARBOHYDRATE AND NITROGENOUS CONSTITUENTS IN THE TERMINAL SHOOTS OF APPLE TREES AND THE EFFECTS OF FIVE VEGETATIVELY PROPAGATED ROOTSTOCKS ON THEM

III. NITROGENOUS CONSTITUENTS

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INTRODUCTION.

THIS paper, the third of the series, reports the results which have been obtained in the section concerning the cycles of nitrogenous materials in samples described fully in Part I (9). From the investigation it is evident that all the principal nitrogen fractions occurred in definite cycles associated with certain seasonal phases and parts of the shoot; but the rootstocks, with the exception of M.IX, did not affect any of the cycles significantly.

HISTORICAL.

Much of the recent work on the nitrogen metabolism of apple trees has been summarized by Karmarkar (3), who studied the effects of two cultural treatments on the chemical composition of the terminal shoots of the variety Newton Wonder worked on Malling II rootstock.

H. L. Colby (1) has since carried out interesting investigations using a number of apple varieties and stocks. He emphasized the following points:

1. Varieties with extremely high nitrogen values showed little or no dwarfing on M.IX and grew only moderately well on M.XIII.

2. The roots of dwarfed trees on M.IX were not starved in respect of starch and nitrogen compounds. The graft unions did not prevent the downward movement of carbohydrate or soluble nitrogen compounds to the roots.

3. Varieties on M.XIII stock continued terminal growth longer than those on M.IX.

Wallace and Warne (11), working on Worcester Pearmain and Lane's Prince Albert, were unable to explain the dwarfing effect of M.IX on the basis of any chemical feature, and the very vigorous rootstocks showed agreement in respect of only one character in the shoots, a high K/N value.

EXPERIMENTAL.

1. FIELD SAMPLING.

Full details of field sampling were presented in Part I (9).

2. PRESERVATION OF SAMPLES.

Preservation of the samples by drying followed the lines described by Karmarkar (3). When dry, the samples were reduced to powder in a Christy and Norris laboratory mill, the final grinding being done by hand until all the material passed through a 60 mesh sieve. Further drying was subsequently necessary to remove moisture absorbed during grinding.

3. METHODS OF ANALYSIS.

In one or two respects, the method of fractionation differed from that employed by Karmarkar (3). Direct extraction of the sample with water was as follows: 30 gram samples of the dry material were weighed out, in duplicate, into cylindrical bottles of 500 ml. capacity. Distilled water, freed from ammonia by long boiling and cooled to room temperature, was then added to the 450 ml. mark, and the bottles were securely corked and agitated in a shaking machine for two hours. The contents were then centrifuged and the extracts removed by decantation. The residual material in the centrifuge tubes was extracted again to give 450 ml. aqueous extract, which was filtered slowly through paper pulp in a Buchner funnel until clear. Soluble proteins were precipitated by bringing to the boiling point after acidifying with 1 ml. glacial acetic acid.

The estimation of nitrate nitrogen by reduction with reduced iron and sulphuric acid was effected in the filtrate from the humin precipitate to avoid hydrolysis of acid amide. Although throughout the experiment a positive quantitative result was almost invariably obtained for nitrate, on no occasion was nitrate detected by qualitative colour tests with diphenylamine hydrochloride or diphenyl benzidine.

The estimation of mono-amino nitrogen by Sörensen's method was not trustworthy, owing to the presence of buffering materials in the solution and some residual colour. The gasometric method of Van Slyke was therefore used. The filtrate from the basic nitrogen precipitate was neutralized by the addition of solid sodium bicarbonate until evolution of CO_2 ceased. After filtration, the solution was concentrated to 10 ml. in an evaporating basin on the water bath.

The amino acid content of the concentrate was determined in the following way: The reaction chamber was of 50 ml. capacity with micro gas burette reading to 0·01 ml. 45 ml. of a 30 per cent. solution of sodium nitrite and 10 ml. glacial acetic acid were shaken together for ten minutes and allowed to displace all the air from the apparatus. Introduction of the extract followed, shaking was continued for a further period of ten minutes, and the gases evolved

were washed twice with alkaline permanganate in the pipette. A "blank" determination was frequently made, and, with the above precautions, duplicate observations agreed closely.

4. Accuracy of the Methods.

Duplicate determinations were made throughout except with stock M.IX, where the samples were very small. Agreement between duplicates was good, especially for the larger groups, total nitrogen, total non-protein nitrogen, imide and basic nitrogen (average divergence of duplicates from the mean, 1·8 per cent.). The smaller fractions, ammonia and nitrate, were usually of the order of less than 10 mg. per cent. residual dry material, and, in consequence, although duplicate titrations rarely differed by more than 0·1 ml. N/50 acid, these data were omitted from the statistical examination.

RESULTS.

Dry weight, as usually estimated in nitrogen studies, includes that of the structural elements of the plant together with reserve materials, mainly carbohydrates; calculations on this basis are thus subject to variation because of the fluctuations which occur in reserve carbohydrates. The residual dry weight, obtained by subtracting reserve carbohydrates (weight of sugars + starch) from the total dry material gives a sounder interpretation of the structural portions of the plant and was therefore chosen as the basis of calculation.

The seasonal cycles of several constituents, expressed as percentages of residual dry weight, are shown graphically in Fig. 1. The values for each fraction are the means for the four rootstocks M.II, M.V, M.VII and M.B. The carbohydrate/nitrogen values are plotted in Figs. 2-7.

DISCUSSION.

1. Seasonal Cycles.

A general similarity to the seasonal cycles observed by Karmarkar (3) in Newton Wonder trees is evident, and consequently, discussion of many of the physiological changes will be omitted. The salient points relating to the seasonal cycles in bark, wood and leaves are described below.

(*a*) *Total water-soluble materials*. The amounts of water-soluble materials in the woody portions of the shoots are small throughout the year, except in the young shoots. In the bark of young shoots the amount is considerable, but it decreases steadily during the summer to reach a minimum in October, followed by a rise in the period of leaf-fall prior to dormancy.

(*b*) *Total Nitrogen*. *Bark*. Bark is always higher in total nitrogen content than wood, except in June. Total nitrogen decreases from a high value in

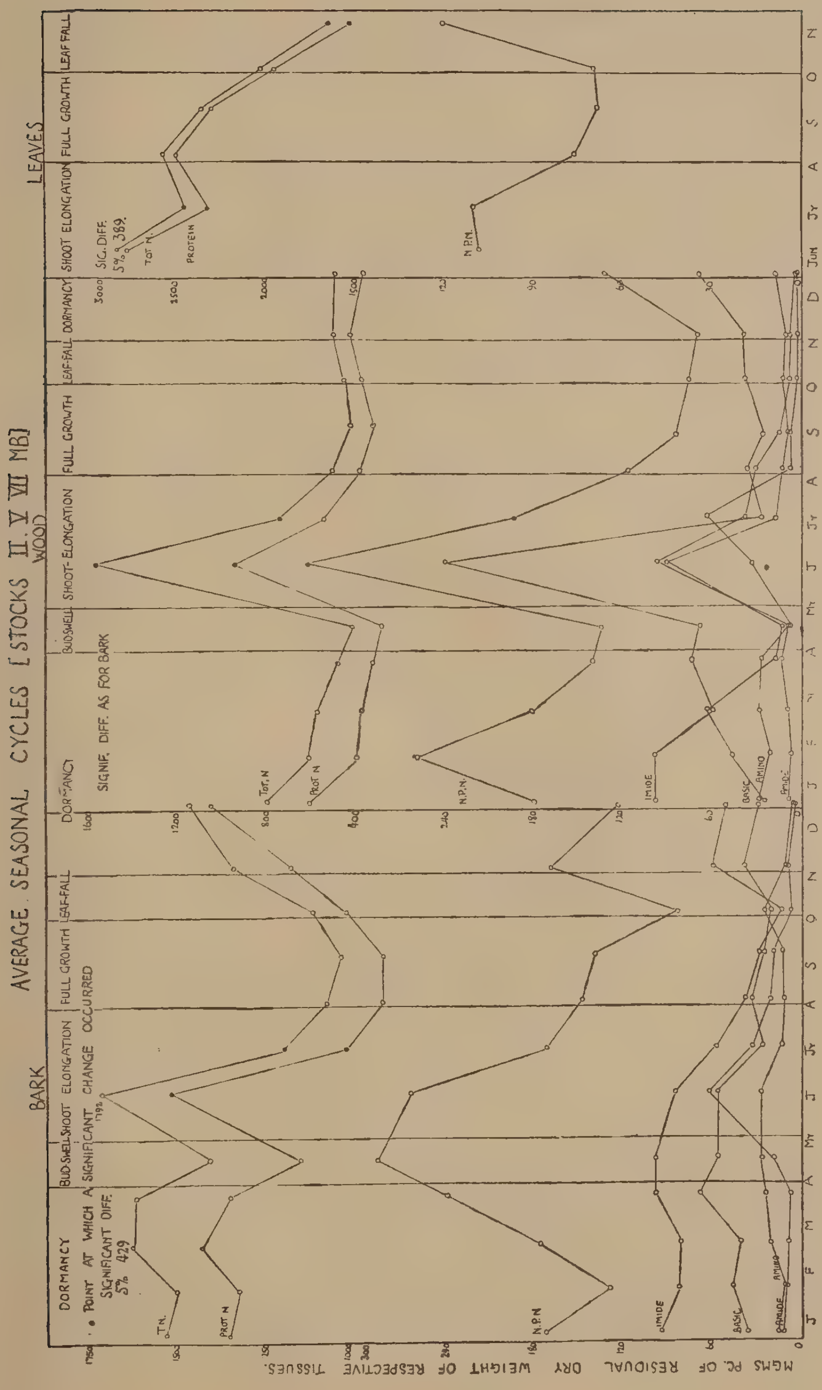

Fig. 1.

Average seasonal cycles for stocks M.II, M.V, M.VII and M.B in bark, wood and leaves.

April to a low one at the time of bud swelling and blossoming. From October until December a continuous rise is shown in the curves, which is specially steep at leaf-fall in November, a phenomenon that appears to be due to migration of nitrogen from the leaves into the bark.

Wood. The young shoots are very rich in nitrogenous constituents, with a prominent peak in June; but, associated with shoot elongation, the total nitrogen content falls rapidly to a minimum value in August.

Leaves. Migration of nitrogen from the leaves to the branches, which has been discussed by many workers, including Combes (2), Thomas (8), Lincoln (4) and Karmarkar (3), is clearly confirmed when the curves for total non-protein

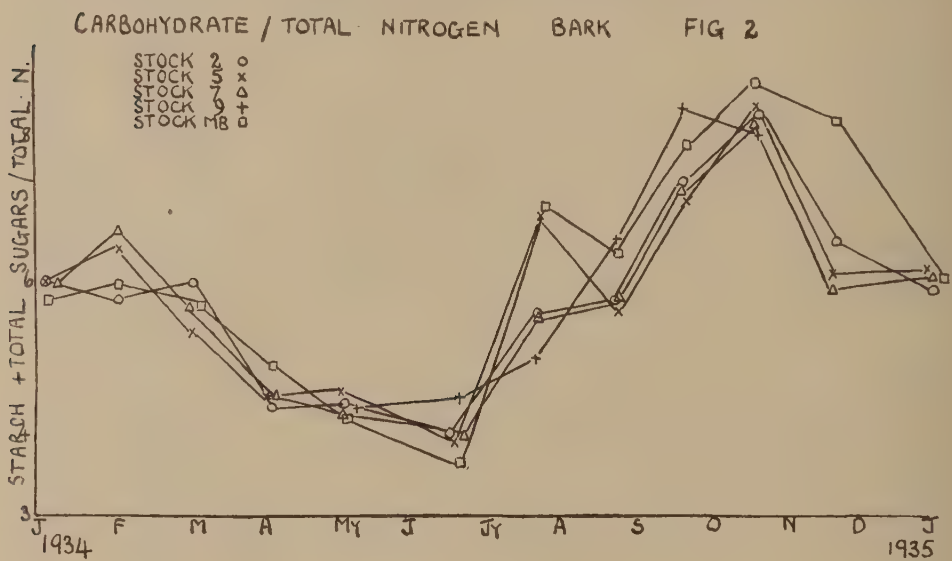

FIG. 2.

nitrogen in bark and leaves are inspected. The quantity of total nitrogen in the leaves, expressed on a residual dry weight basis, is highest at the beginning of the season, and, except for a slight halt in August, falls continuously until defoliation in November, at a rate scarcely accounted for by the rapid growth of the leaves.

(*c*) *Total non-protein nitrogen.* In bark and leaves the maximum amount of non-protein nitrogen occurs during the bud swelling period with a minimum in October at the cessation of growth.

In the woody tissues, this constituent is fairly constant throughout the year except in the young shoots in June, which contain very large quantities of non-protein nitrogen.

(*d*) *Reserve carbohydrate/nitrogen relationships.*

Reserve carbohydrates/total nitrogen. Bark (*Fig.* 2). The ratio carbohydrate/total nitrogen falls continuously from dormancy until June, and afterwards the value rises sharply till the time of defoliation. There is a subsequent decline at the approach of dormancy, presumably due to the continuous accumulation of nitrogen at that period.

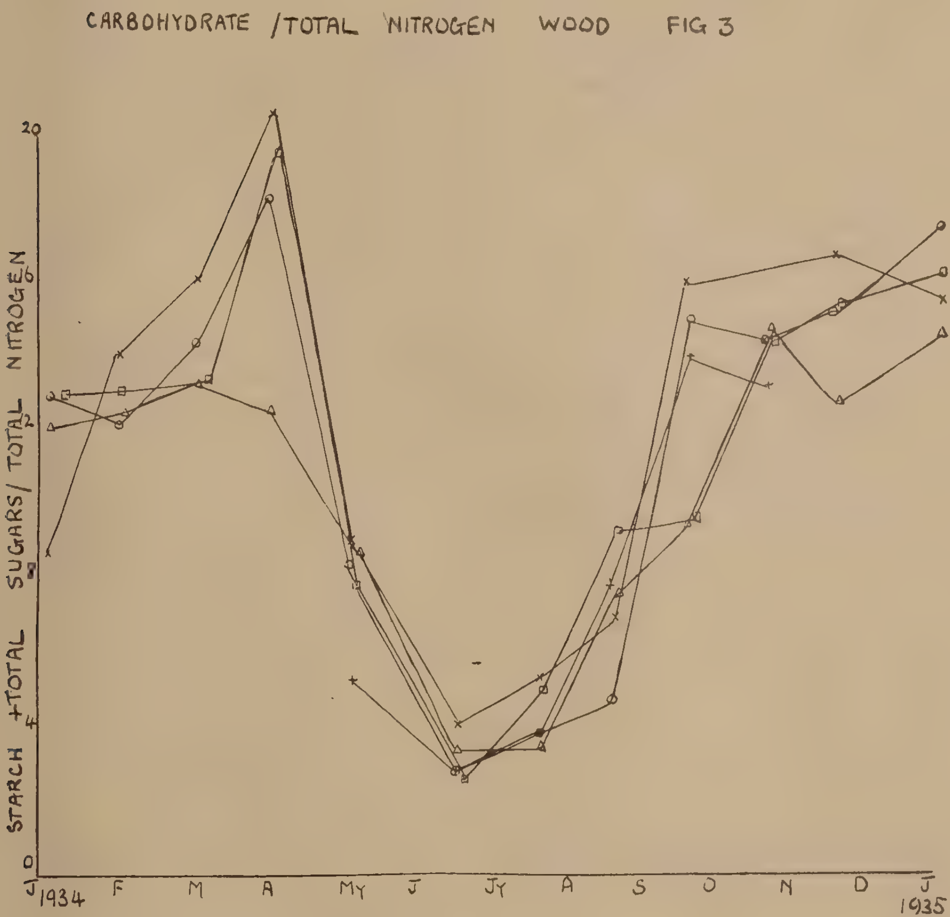

FIG. 3.

Wood (*Fig.* 3). There is a protracted rise in carbohydrate/nitrogen from dormancy until bud break in April, followed by a fall in July, when the young shoots are growing rapidly with consumption of reserve carbohydrates. From July onwards a gradual replenishment of carbohydrate reserves takes place with ageing of the shoots.

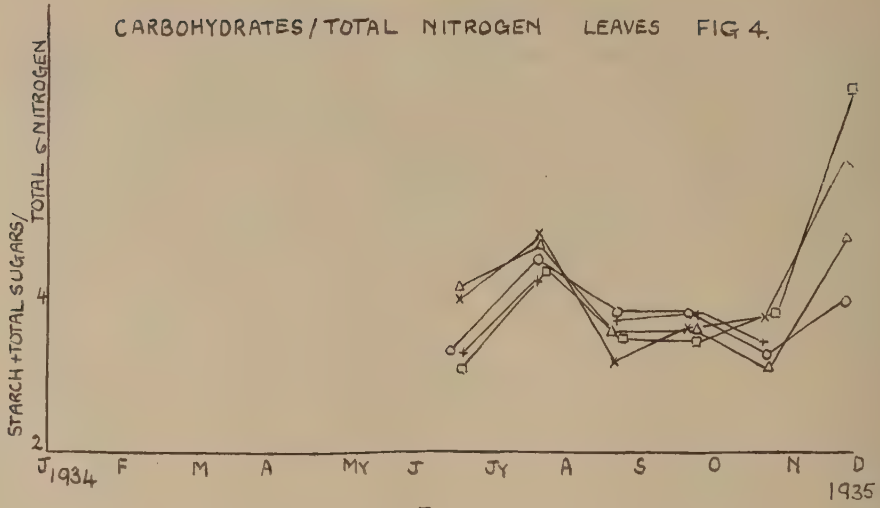

FIG. 4.

Leaves (*Fig.* 4). The ratio is constant throughout the seasonal cycle with the exception of peak values in July and at defoliation, the former peak possibly being associated with a period of high assimilation of carbohydrate and the latter with removal of nitrogen from the leaves to the bark.

Reserve carbohydrates / total non-protein nitrogen (*Figs.* 5-7). The curves for this ratio in bark and wood are very similar to those for reserve carbohydrates / total nitrogen. The parallelism does not hold for leaves, however,

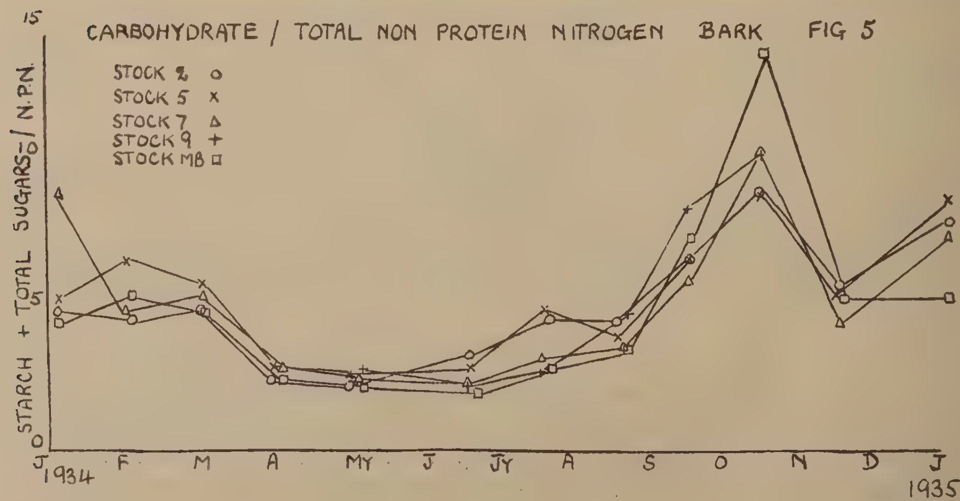

FIG. 5.

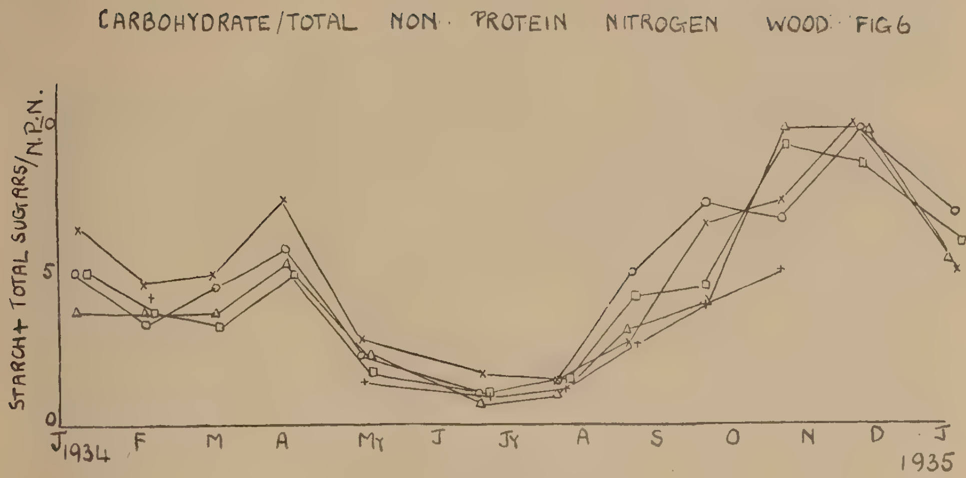

FIG. 6.

where the values are low at the beginning and the end of foliation, and high during the intermediate period.

(*e*) *Nitrate nitrogen. Bark and Wood.* The highest figures recorded for nitrate nitrogen in bark and wood, according to the quantitative method employed, were 11 and 9·5 mg. per cent. residual dry weight respectively. At no time, however, was a positive qualitative test for nitrate nitrogen given with diphenylamine or diphenyl benizidine. Apparently, all nitrate was reduced

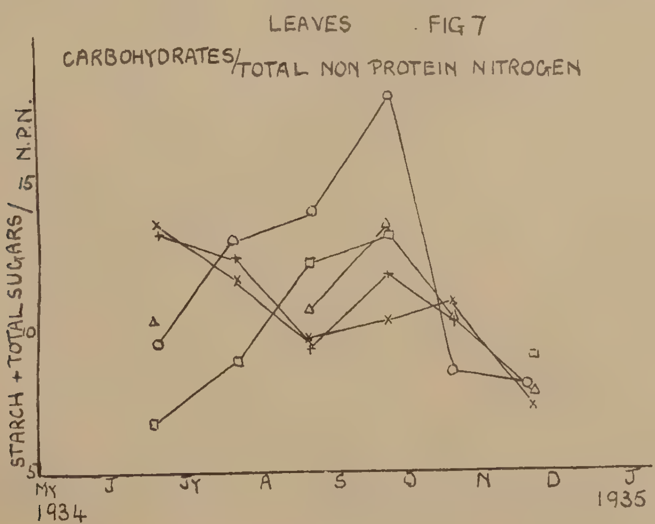

FIG. 7.

and elaborated to organic form before reaching the shoots, a conclusion in harmony with the results of many other experiments (Stuart (7)).

(*f*) *Ammonia nitrogen. Bark and Wood.* The quantities of ammonia in the shoots are always small, the highest recorded values for bark and wood being 15 and 7 per cent. of the total non-protein nitrogen respectively.

Ammonia nitrogen reaches a high level in the bark during the bud swelling period, and again in the months of August and September. In the woody portion of the shoot, ammonia nitrogen is appreciable only in June, a time of great activity in the plant.

(*g*) *Acid amide nitrogen.* In view of the current conceptions of the functions of acid amide in plants it is of interest to consider the curves for this fraction in relation to those for ammonia, and in connection with carbohydrate reserves.

In bark, ammonia accumulates in May and in August with minimum values during the period June and July. Acid amide, on the other hand, occurs in very low amounts in April, when the quantities of ammonia are appreciable, and is present in considerable quantities during the whole of the growing season, with a maximum in June when little ammonia is formed. The reasons for peak values for acid amide in June accompanied by a low value for ammonia are not clear, since supplies of reserve carbohydrates at that time are low in the shoots. Moreover, high values for ammonia would scarcely be expected in the presence of high carbohydrate reserves from August to November.

In the wood, during the active spring period, ammonia and acid amide run parallel to one another. After June, however, ammonia falls to a negligible level, whereas acid amide declines only gradually, possibly as a result of more plentiful supplies of carbohydrate.

(*h*) *Humin nitrogen.* Expressed on a percentage non-protein nitrogen basis, humin nitrogen is approximately constant throughout the year at 15 per cent. for bark and 10 per cent. for wood. The seasonal changes in mono- and di-amino acids are similar to those observed for humin, in agreement with previous work on the origin of the humin fraction.

Karmarkar (3) pointed out the similarity in the cycles of humin and total water-soluble materials, of which carbohydrates usually form a considerable part. However, although the humin fraction in wood appears to be closely associated with the amounts of water-soluble materials, notable differences occur in bark, especially during the bud swelling period.

Recently, Orcutt and Wilson (6), assuming the humin condensation to be oxidative in nature, have eliminated this confusing loss in fractionation by reduction with sodium bisulphite.

(*i*) *Basic nitrogen*. In both bark and wood, the percentage of basic nitrogen in total non-protein nitrogen varies little, though there are tendencies for high values to occur just preceding new growth (Mulay (5)) with minima during the active growing period from June until August. Karmarkar (3) reported a minimum in August, which may be due to utilization of basic nitrogen compounds in the synthesis of nucleo-proteins.

(*j*) *Imide nitrogen*. The curves are all similar to those described in the grass and arable investigation of Karmarkar. In bark, imide decreases continuously from January until October, with accumulation from the latter month until January. From the chemical nature of the imide fraction it would be expected that these bodies would predominate in the dormancy period, but in the woody portion of the shoots values of 50-60 per cent. non-protein nitrogen are recorded in the spring.

(*k*) *Mono-amino acid nitrogen*. Stuart (7) has described the interference of tannins in the Van Slyke estimation of amino nitrogen. In the present investigation, this difficulty was not encountered since such di- and tri-hydric phenolic bodies were precipitated or denatured on distillation with magnesium oxide in the acid amide determination.

In bark, maxima occur during the period of active nitrogen mobilization in spring and just prior to defoliation, during considerable changes in nitrogenous constituents.

As percentage of non-protein nitrogen, high levels are present in woody tissues during the early spring, followed by a minimum in May and June, and then a sharp peak during July.

(*l*) *Rest nitrogen*. By the systematic analysis employed, it was possible to account for at least 95 per cent. of the total non-protein nitrogen and, in most samples, "rest" nitrogen was not appreciable.

2. STOCK DIFFERENCES.

The morphological differences have been described in Part I (9), and chemical data alone are discussed here.

Statistical analysis of these data shows that the nitrogenous fractions in bark and wood are strikingly similar for all five rootstocks.

It is necessary to stress the point that all chemical data (with the exception of those concerning nitrate and ammonia) were included in the statistical examination, the highest fractions, such as total nitrogen of the order of 2,000 mg. per cent. residual dry material, and the lowest, acid amide, of the order of 10 mg. per cent. This appeared to be the most satisfactory way of treating the results, but, on this basis, the large remainder from the high

individuals negatives any variation of the lower fractions, and these have been examined separately.

In the woody portions of the shoots, the total nitrogen of stock M.IX was significantly in excess of that in all the other rootstocks, which were not significantly different from one another.

Stock M.IX seems to be associated with high acid amide content in the scion, especially in the young shoots. Asparagin is generally assumed to be the storage form of ammonia, and its formation in quantity appears to be characteristic of this precocious rootstock.

No relationships are apparent in the chemical data similar to the gradation of pomological properties, and rootstock M.IX alone induces any significant change in the chemical composition of long established trees.

The nitrogen cycles for Lane's Prince Albert closely resemble those obtained previously at Long Ashton in the Newton Wonder experiment, and a similarity has also been observed by Smyth in the carbohydrate cycles (Part II (10)).

3. Double Interaction of Factors.

Tables IA and IB, IIA and IIB show the analysis of variance data for bark, wood and leaves, and the several interactions of different factors are discussed below.

(1) *Stocks and Months.* The variation due to the interaction between all the rootstocks, M.II, M.V, M.VII, M.IX and M.B, and months is not significant in bark and wood. The cycles are similar in trend, and monthly variations are the same for all the rootstocks. In the leaves, however, all the stocks have significant interactions of stocks and months, and the seasonal cycles are not parallel. Thus, monthly changes in the leaves of the scion are affected by the rootstock employed.

Rootstock M.IX has a cycle in bark and wood coincident with those on the other stocks, although its growth responses are different.

(2) *Stocks and Nitrogen Fractions.* The variation due to the interaction between stocks and nitrogen fractions in bark, wood and leaves is not significant; thus, partition into nitrogen fractions is independent of the rootstock.

(3) *Months and Nitrogen Fractions.* The variation due to the interaction of months and nitrogen fractions in bark, wood and leaves is significant. From one month to another the changes in the fractions are not the same, the differences being most noticeable for the larger fractions total nitrogen, protein nitrogen and total non-protein nitrogen.

(4) *Stocks and Parts of the Plant.* The interaction between stocks and bark and wood portions is significant only for stock M.IX, which means that the

Table Ia.

Showing Analysis of Variance of Percentage of Nitrogen Fractions in Bark and Wood Portions of Shoots—Stocks M.II, M.V, M.VII and M.B.

Results of 12 months' Sampling.

Source of Variation.	Degrees of Freedom.	Sum of Squares.	Mean Square.	$\log_e$.	$\frac{1}{2}$ ($\log_e$ Factor—$\log_e$ Error).	1% Points.	5% Points.	Remarks.
Between means of Months ..	11	33475841·8	3043258·3	14·9284	1·9509	0·3908	0·2804	Significant.
,, ,, Stocks ..	3	283905·6	94635·2	11·4578	0·2156	0·6651	0·4787	Not significant.
,, ,, Nitrogen fractions	8	1169431041·6	146178880·2	18·8005	3·8869	0·4604	0·3309	Significant.
,, ,, Parts of plant	1	63197444·6	63197444·6	17·9618	3·4676	0·9462	0·6729	Significant.
Interactions: Months—Stocks ..	33	1454840·2	44086·1	10·6939	—	0·2913	0·2085	Not significant.
Months—N fractions ..	88	53474247·2	607661·9	13·3175	1·1455	0·0000	0·0000	Significant.
Months—Parts of Plant ..	11	9520279·2	865479·8	13·6711	1·3223	0·3908	0·2804	Significant.
Stocks—Parts of Plant ..	3	263095·4	87698·5	11·3816	0·1775	0·6651	0·4787	Not significant.
Stocks—N fractions ..	24	622968·4	25957·0	10·1644	—	0·2913	0·2085	Not significant.
Parts of Plant—N fractions	8	215144772·2	26893096·5	17·1073	3·0404	0·4604	0·3309	Significant.
Remainder ..	673	41383042·4	61490·4	11·0266				
Total ..	863	1588251478·6						

TABLE IB.

Showing Analysis of Variance of Percentage of Nitrogen Fractions in Bark and Wood Portions of Shoots—Stocks M.II, M.V, M.VII, M.IX and M.B.

Results of 7 months' Sampling.

Source of Variation.	Degrees of Freedom.	Sum of Squares.	Mean Square.	$\log_e$.	½ ($\log_e$ Factor—$\log_e$ Error).	1% Points.	5% Points.	Remarks.
Between means of Months ..	6	39871374·3	6645229·1	15·7095	2·4025	0·5152	0·3706	Significant.
,, ,, Stocks ..	4	695938·7	173984·7	12·0668	0·5812	0·5999	0·4319	Significant.
,, ,, Nitrogen fractions	8	821147062·8	102643382·9	18·4462	3·7709	0·4604	0·3309	Significant.
,, ,, Parts of plant	1	25434426·7	25434426·7	17·0505	3·0736	0·9462	0·6729	Significant.
INTERACTIONS : Months—Stocks ..	24	1955237·3	81468·2	11·3080	0·2019	0·2913	0·2085	Not significant.
Months—N fractions ..	48	59793160·6	1245690·9	14·0354	1·5656	0·0000	0·0000	Significant.
Months—Parts of Plant ..	6	7643292·7	1273882·1	14·0577	1·5767	0·5152	0·3706	Significant.
Stocks—Parts of Plant ..	4	1183698·4	295924·6	12·5978	0·8468	0·5999	0·4319	Significant.
Stocks—N fractions ..	32	1196185·2	37380·8	10·5288	—	0·2913	0·2085	Not significant.
Parts of Plant—N fractions	8	101959931·2	12744991·4	16·3603	2·7280	0·4604	0·3309	Significant.
REMAINDER ..	488	26553214·1	54412·3	10·9043				
TOTAL ..	629	1087433522·0						

TABLE IIA.

Showing Analysis of Variance of Percentage of Nitrogen Fractions in Leaves—Stocks M.II, M.V, M.VII and M.B.

Results of 6 months' Sampling.

Source of Variation.	Degrees of Freedom.	Sum of Squares.	Mean Square.	$\log_e$.	$\frac{1}{2}$ ($\log_e$ Factor—$\log_e$ Error).	1% Points.	5% Points.	Remarks.
Between means of Months ..	5	52442482·8	10488496·6	16·1659	3·1612	0·6540	0·4648	Significant.
,, ,, Stocks ..	3	566681·7	188893·9	12·1491	1·1528	0·7531	0·5362	Significant.
,, ,, Nitrogen fractions	2	775068468·6	387534234·3	19·7753	4·9659	0·8423	0·5994	Significant.
INTERACTIONS :								
Months—Stocks ..	15	3032221·6	202148·1	12·2165	1·1865	0·5224	0·3691	Significant.
Months—N fractions ..	10	26812275·7	2681227·6	14·8017	2·4791	0·5773	0·4090	Significant.
Stocks—N fractions ..	6	431795·0	71965·8	11·1840	0·6702	0·6226	0·4420	Hardly significant.
REMAINDER ..	30	1536048·3	51201·6	9·8435				
TOTAL ..	71	859889973·7						

TABLE IIB.

*Showing Analysis of Variance of Percentage of Nitrogen Fractions in Leaves—
Stocks M.II, M.V, M.VII, M.IX and M.B.*

Results of 5 months' Sampling.

Source of Variation.	Degrees of Freedom.	Sum of Squares.	Mean Square.	$\log_e$.	$\frac{1}{2}$ ($\log_e$ Factor—$\log_e$ Error).	1% Points.	5% Points.	Remarks.
Between means of Months ..	4	28095482·7	7023870·7	15·7647	2·3908	0·6954	0·4947	Significant.
,, ,, Stocks ..	4	619525·4	154881·4	11·9505	0·4837	0·6954	0·4947	Significant.
,, ,, Nitrogen fractions	2	925237861·7	462618930·8	19·9524	4·4846	0·8423	0·5997	Significant.
INTERACTIONS :								
Months—Stocks ..	16	3144405·3	196525·3	12·1884	0·6026	0·5224	0·3691	Significant.
Months—N fractions ..	8	12529494·3	1566186·8	14·2641	1·6405	0·5773	0·3702	Significant.
Stocks—N fractions ..	8	393267·4	49158·4	10·8028	0·0902	0·5773	0·3702	Not significant.
REMAINDER ..	32	1884009·0	58875·3	10·9832				
TOTAL ..	74	971904045·8						

ratio bark/wood for the sum of all the nitrogen fractions is the same for all the rootstocks except M.IX.

(5) *Nitrogen Fractions and Parts of the Plant.* The variation due to the interaction between nitrogen fractions and parts of the plant is significant for all stocks. Different parts of the plant, therefore, do not contain proportionate amounts of the various nitrogen groups.

(6) *Months and Parts of the Plant.* The variation due to the interaction between months and bark and wood is significant. The seasonal cycles for different nitrogen fractions in bark differ significantly from those in wood, and separation of these portions in shoots is therefore essential.

CONCLUSIONS.

1. The seasonal cycles of nitrogenous materials in Lane's Prince Albert trees on the five rootstocks used exhibit a marked similarity to those described for Newton Wonder trees under two cultural systems, grass and arable.

2. The seasonal cycles of nitrogenous materials in shoots grown on rootstock M.IX are different from those in shoots on the other stocks, M.II, M.V, M.VII and M.B, which produce no significant differences in the scion variety. Although the only significant difference shown by M.IX is for the value of total nitrogen in wood, a marked tendency was observed in the trees on this rootstock to accumulate high amounts of acid amide.

3. The rootstocks produced differences in pomological character of the scion which were not correlated with the nitrogen content of the terminal shoots.

4. Statistical analysis leads to the following further conclusions:

(1) The total concentration of the various nitrogen constituents is independent of the rootstock.

(2) The proportions of the constituents present in bark as compared with the amounts in wood are alike for all the rootstocks except stock M.IX.

(3) The seasonal cycles of the various nitrogen fractions differ significantly from each other, the differences being most noticeable for the larger fractions total nitrogen, protein nitrogen and total non-protein nitrogen.

(4) The seasonal cycles in bark differ significantly from those in wood.

(5) The amounts of individual nitrogen fractions are different in bark and wood portions of the shoot.

SUMMARY.

1. The seasonal cycles of nitrogenous materials in the terminal shoot portions of Lane's Prince Albert apple, grafted on five Malling stocks, M.II, M.V, M.VII, M.IX and M.B, have been determined. The objects of the experiment were (1) to obtain evidence on the effect of the various stocks on the chemical composition of the shoots and (2) to compare the results with the previous work of Karmarkar on nitrogen metabolism in Newton Wonder apple trees.

2. Samples were collected at monthly intervals from January 1934 until January 1935 from eight trees on each rootstock. The shoots were separated into the portions bark, wood and leaves.

3. The chemical procedure was as described by Karmarkar, with the exception of the Van Slyke estimation of amino acid. Total nitrogen, total non-protein nitrogen, ammonia, acid amide, humin, basic, imide, mono-amino and proteose nitrogen were determined.

4. The cycles were similar in trend to those described in a previous study for Newton Wonder on M.II.

5. Rootstock M.IX produced in the scion variety significantly high total nitrogen in wood, and a tendency to accumulation of acid amide ; but no significant differences were observed between the composition of shoots grown on the other rootstocks, M.II, M.V, M.VII and M.B.

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STUDIES ON THE TOXICITY OF CERTAIN NITROPHENOLS, THIOCYANATES, NAPHTHALENE DERIVATIVES AND ORGANIC BASES TO THE EGGS OF SOME COMMON ORCHARD PESTS

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INTRODUCTION.

The differences in ovicidal action between tar distillates and petroleum oils have necessitated the use of both types as winter washes for fruit trees. Tar distillates are toxic to the eggs of Aphides and Apple Sucker but have little effect on those of Capsids or Red Spider. Petroleum oils, on the other hand, are toxic to the eggs of Capsids and Red Spider but not to those of Aphides and Apple Sucker. The advantage of controlling both groups of pests by a single wash is so obvious that considerable attention has been given to devising one. A direct result was the introduction of the tar-petroleum mixed oil preparations. These, however, are liable to damage the buds and can be used only during the fully dormant period. Moreover their cost is high. A dual purpose wash is, therefore, still needed. Suggested possibilities are (a) the inclusion in the petroleum wash of some substance toxic to the eggs of Aphides and Apple Sucker and (b) the discovery of a single substance toxic to both types of egg. These problems have been intensively studied at East Malling for the past four years.

The materials tested included:—

- Dinitrophenols and their salts.
- Aliphatic thiocyanates.
- Naphthalene and some derivatives ; certain allied substances.
- Pure bases known to be present in coal tar ; related substances ; alkaloids.
- Crude tar acids.
- Crude tar bases.

The crude tar acids and bases are not considered here, but will be dealt with in a later paper on hydrocarbon oils.

The potassium salt of 3:5-dinitro-*o*-cresol was used as an insecticide as early as 1892. It was, however, Gimingham and Tattersfield and their colleagues who pointed out the value of the nitrophenols as ovicides. They

reported (30, 8) the high toxicity under laboratory conditions of 3:5-dinitro-*o*-cresol and its salts to eggs of *Selenia tetralunaria* Hufn. and demonstrated (8) under field conditions its efficacy against Hop-Damson Aphis (*Phorodon humuli* Schr.). Gimingham and Tattersfield (9) showed that the dinitrocresol and its sodium salt were each toxic in the laboratory to the eggs of Winter Moth (*Operophtera brumata* L.) and Vapourer Moth (*Orgyia antiqua* L.) among others, but that they were apparently without effect on those of Red Spider (?*Oligonychus ulmi* C. L. Koch). In the field Green Apple Aphis (*Aphis pomi* De Geer), Rosy Apple Aphis (*Anuraphis roseus* Baker) and Apple Sucker (*Psyllia mali* Schmidt) were controlled and caterpillars greatly reduced. Extensive field trials reviewed by the same authors (10) confirmed these results.

Despite these results, and possibly because of difficulties in compounding and applying the preparations, little more was heard of the nitrophenols as ovicides until Kagy and Richardson (13), in 1936, noted the toxicity of a solution of 2:4-dinitro-6-*cyclo*hexylphenol in petroleum oil to *Lygaeus kalmii* Stål., and Dutton (5) obtained good control of Rosy Apple Aphis in the field by the same mixture. By that time experiments had been in progress for two years at East Malling, following up those of Gimingham and Tattersfield. The aim was to study the toxicity of the dinitrophenols by the more highly standardized laboratory methods now available (29) and to try to overcome the difficulties of preparation and application of washes containing them.

Attention was concentrated upon the free nitrocresol because it is soluble in petroleum oil and causes less staining than the sodium salt. The low solubility (about 2 per cent.) of 3:5-dinitro-*o*-cresol in Grade E (20) petroleum oil was at first considered a disadvantage. Toxicity tests, however, indicated that the amount of nitrocresol soluble in the 5-6 per cent. of petroleum oil normally applied was sufficient to control Aphides.

The thiocyanates have only recently been examined as potential ovicides although they have attracted much attention as contact insecticides. Kearns and Martin (15), following up the observations of Murphy and Peet (24) on the ovicidal action of thiocyanates against Citrus Mealy Bug (*Pseudococcus citri* Risso), were the first to report on the possible use of this type of compound instead of tar oil. Lauryl rhodanate (dodecyl thiocyanate) was found to be highly toxic, much more so than cetyl thiocyanate, to eggs of *A. pomi*, and gave promising results in a preliminary field trial. Jary and Austin (12) have recently shown that washes containing either dodecyl thiocyanate or β-butoxy-β'-thiocyanodiethyl ether (23) are effective against eggs of the Hop Red Spider (*Tetranychus telarius* L.). The present paper records the results of experiments with the above-mentioned thiocyanates, with the separate members of the C_6 to C_{12} series of *secondary* thiocyanates and with a miscellaneous group of other aliphatic thiocyanates.

Each member of the naphthalene group has been studied by at least one previous investigator. Tattersfield *et al.* (30) found that naphthalene and decalin had little effect on eggs of *S. tetralunaria* whereas tetralin and α-chlornaphthalene, especially the latter, were highly toxic. On the other hand, Kearns *et al.* (16) showed that tetralin was only slightly toxic to eggs of *A. pomi,* as were also decalin and anthracene. A mixture of polychlornaphthalenes, known as Halowax, has been studied by Breakey. He reported (2) a high toxicity to eggs of *T. telarius,* while Breakey and Miller (3, 4) found this material to be toxic to eggs of the Angoumois Grain Moth, a Blow Fly, the Oriental Fruit Moth and the Codling Moth. The present authors have tested a series of chlornaphthalenes, ranging from α-monochlornaphthalene (21·8 per cent. Cl) to a polychlornaphthalene (Seekay Wax) containing 56·0 per cent. Cl. Several other compounds, not strictly derivatives of naphthalene, are for convenience considered with this group. Benzene was necessarily tested as it was used as a solvent in these tests. Staniland *et al.* stated (28) that "diphenyl oxide" (?diphenylene oxide) was highly toxic to eggs of Winter Moth, and Kearns *et al.* reported (16) that "diphenyl oxide (dibenzofuran)" had little or no toxic action on the eggs of *A. pomi.* In view of the somewhat ambiguous nomenclature of the substances used by the above workers, both diphenyl oxide (diphenyl ether) and diphenylene oxide (dibenzofuran) were used in the present authors' tests on eggs of Winter Moth.

The authors' experience with the crude tar bases from tar distillate oils led them to test the ovicidal properties of individual bases known to be present in tar, and of closely related bases. It was convenient to test with them nicotine and anabasine, both derivatives of pyridine. Their interest in the ovicidal properties of nicotine began in 1933 when Hey and Steer (11) concluded from the results of field spraying trials that nicotine is toxic to the egg of the Apple Sawfly (*Hoplocampa testudinea* Klug.) at any stage of its development. Many other authors have found nicotine to be toxic to insect eggs, especially those of Aphides and of various species of Lepidoptera. Very little work with the other bases is on record, but Moore and Graham (22) found quinoline to be more toxic than pyridine to eggs of the Colorado Beetle.

LABORATORY TESTS.

Methods and Materials.

Chemical.

All the substances were made up in 1 per cent. soap solutions, i.e. 1 per cent. oleic acid plus 0·128 per cent. sodium hydroxide. The nitrophenols were also used in sulphite-lye emulsions of petroleum oils, and the chlornaphthalenes in casein-Turkey Red oil or goulac emulsions. All the preparations listed in

the tables of results were, unless otherwise stated, in soap solutions. The soap emulsions were prepared by the usual two-solution process. With liquids, the material was mixed with oleic acid and added to the requisite amount of sodium hydroxide dissolved in distilled water. All quantities here are volume in volume. All the thiocyanates, α-monochlornaphthalene, tetralin, decalin, benzene, and all the bases except 2:6-dimethylquinoline and carbazole, were prepared thus and gave satisfactory emulsions or, in some cases, solutions. Solids were dissolved in an appropriate solvent (weight in volume), indicated in brackets in the tables of results, and the solution emulsified as above. Benzene, tetralin and decalin were used as solvents, the first named being by far the most satisfactory. A description of all the substances used is given in Table I. All the physical constants are taken from the literature or from private communications.

Biological.

The methods used have already been described by one of us (29). Most of the work was done on eggs of the Vapourer Moth (*Orgyia antiqua*), the Green Apple Aphis (*Aphis pomi*) and the Winter Moth (*Operophtera brumata*), but some tests have been made on eggs of Red Spider (*Oligonychus ulmi*) and Apple Sucker (*Psyllia mali*).

In the Tables of Results the first column shows the number of each test and the season in which it was carried out. Thus 67/35 represents test number 67 of the 1935-36 season.

Results.

Vapourer Moth.

The viability of Vapourer Moth eggs and their susceptibility to toxic materials varies a great deal from batch to batch. Hence critical methods of statistical analysis cannot well be applied except to individual tests, in which the performance of several materials can be measured on the same batch of eggs. For purposes of comparison, therefore, for each treatment and for each egg batch a "per cent. hatch" has been calculated as

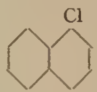
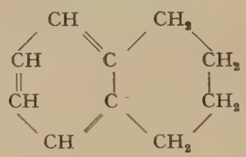
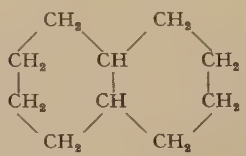
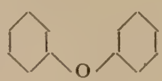
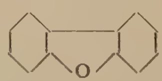
$$\frac{\text{per cent. hatch of treated eggs} \times 100}{\text{per cent. hatch of similar untreated eggs.}}$$

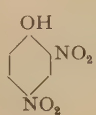
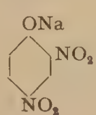
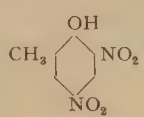
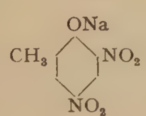
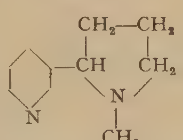
This gives the same value (100 per cent.) to the hatch in each of the many control series of untreated eggs.

In Tables II-V, the figures in column 6 indicate the number of replications used to obtain the estimate of the mean hatch and its standard error (columns 4 and 5), whilst those in column 7 show the total number of eggs from which the data were derived.

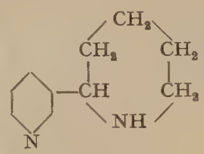
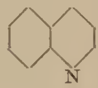
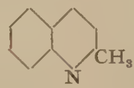
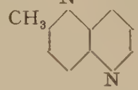
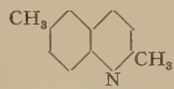
TABLE I.

Materials Employed.

Naphthalene			pure	m.p. 80° C.; b.p. 218° C.
α-Monochlornaphthalene ..		21·8% Cl.	commercially pure	b.p. 250-2° C.
Polychlornaphthalenes:				
Chlorinated naphthalene R.77		c. ?46·8% Cl.	commercially refined	m.p. c. 77° C.
Seekay Wax R.68 ..		c. 46·0% Cl.	„	m.p. c. 68° C.
„ „ R.93 ..		c. 50·5% Cl.	„	m.p. c. 93° C.
„ „ R.113 ..		c. 54·0% Cl.	„	m.p. c. 113° C.
„ „ R.123 ..		c. 56·0% Cl.	„	m.p. c. 123° C.
Tetralin			commercially pure	b.p. 206° C.
Decalin			„	b.p. 187-8° C.
Thionaphthene			pure	m.p. 30-1° C.; b.p. 220-1° C.
Diphenyl oxide			„	m.p. 28° C.; b.p. 253° C.
Diphenylene oxide ..			„	m.p. 81° C.; b.p. 288° C.
Benzene			„	b.p. 80° C.

Anthracene		pure	m.p. 217° C.; b.p. 351° C.
Lauryl rhodanate* / *n*-Dodecyl thiocyanate*	$C_{12}H_{25}$. SCN	technical / commercially pure	b.p. 170-2°C/10mm.
Cetyl " ..	$C_{16}H_{33}$. SCN	technical	b.p. 222-7°C/13mm.
sec.-Hexyl " ..	C_4H_9. CH(CH_3). SCN	commercially pure	b.p. *c.* 65° C./4mm.
" Heptyl " ..	C_5H_{11}. CH(CH_3). SCN	"	b.p. *c.* 80° C./4mm.
" Octyl " ..	C_6H_{13}. CH(CH_3). SCN	"	b.p. *c.* 95 °C./4mm.
" Nonyl " ..	C_7H_{15}. CH(CH_3). SCN	"	b.p. *c.* 110°C./4mm.
" Decyl " ..	C_8H_{17}. CH(CH_3). SCN	"	b.p. *c.* 125°C./4mm.
" Undecyl " ..	C_9H_{19}. CH(CH_3). SCN	"	b.p. *c.*140° C./4mm.
" Dodecyl " ..	$C_{10}H_{21}$. CH(CH_3). SCN	"	b.p. *c.*155° C/4mm.
" C_{10}—C_{12} " ..		"	
β-Butoxy-*β'*-thiocyano-diethyl ether	C_4H_7O. $(CH_2)_2$. O. $(CH_2)_2$. SCN	75% in kerosene	
Methylallyl thiocyanate ..	CH_2:C(CH_3)CH_2. SCN	commercially pure	
tert.-Butyl thiocyanate ..	$(CH_3)_3$. C. SCN	"	
2:4-Dinitrophenol		pure	m.p. 114-15° C.
Sodium 2:4-dinitrophenate		"	
3:5-Dinitro-*o*-cresol ..		"	m.p. 86-7° C.
Sodium 3:5-dinitro-*o*-cresylate		"	
Nicotine		commercial 95/98%	b.p. 246° C./ 730 mm.

* Lauryl rhodanate is the technical grade of dodecyl thiocyanate.

Anabasine		bases from crude sulphate	b.p. 276° C.
Pyridine		pure	b.p. 114° C.
Quinoline		„	b.p. 240° C.
2-Methylquinoline (Quinaldine) ..		„	b.p. 247° C.
6-Methylquinoline ..		„	b.p. 258° C.
2:6-Dimethylquinoline ..		„	m.p. 60° C.; b.p. 266-7° C.
iso-Quinoline		„	b.p. 240° C.
Pyrrole		„	b.p. 130-1° C.
Carbazole		„	m.p. 238° C.; b.p. 354-5° C.

	Neutral oil % by weight.	Sp. gr.	Neutral oil.				
			Boiling range			Unsul-phonated residue % by by volume.	Soluble in dimethyl sulphate % by volume.
			10%	50% by volume.	80%		
Petroleum oil A (a spindle oil)	100·0	0·901	347° C.	368° C.	389° C.	72	4
Petroleum oil B (an Endeleanu extract)	97·2	0·997	321° C.	357° C.	390° C.	16	22

TABLE II.

Toxicity of Dinitrophenols and their Salts to O. antiqua *eggs.*

Test No.	Material tested.	Conc. %	Hatch % Controls = 100	S.E.	n	Total eggs.
33/34	Sodium 2:4-dinitrophenate	1·0	34·5	4·9	2	167
66/35	„ „	0·5	82·0	19·9	6	334
„	„ „	1·0	56·1	12·7	6	435
„	„ „	1·5	36·9	16·7	6	432
33/34	Sodium 3:5-dinitro-*o*-cresylate ..	1·0	14·0	14·0	2	168
67/35	„ „	0·5	29·8	11·4	6	354
„	„ „	1·0	6·7	3·2	6	394
„	„ „	1·5	5·7	1·9	6	365
32/36	„ „	0·25	17·7	9·3	6	502
„	„ „	0·5	0	—	6	545
33/36	2:4-Dinitrophenol * (in 5% acetone + 10% benzene)	0·5	10·6	3·6	5	437
„	„ („ „)	1·0	7·3	4·5	5	403
32/36	3:5-Dinitro-*o*-cresol * (in 10% benzene)	0·5	0	—	6	521
33/36	„ („ „)	1·0	0	—	5	382
57/35	Benzene	20·0	105·8	5·3	4	354
12/34	Controls (sodium oleate)	1·0	103·4	6·1	6	400
41/35	„ „	1·0	92·7	4·8	6	305

* Probably present to some extent as sodium salt as a result of the process of emulsification. The emulsion of nitrophenol was rather unsatisfactory.

TABLE III.

Toxicity of Thiocyanates to O. antiqua *eggs.*

Test No.	Material tested.	Conc. %	Hatch % Controls = 100	S.E.	n	Total eggs.
87/35	Lauryl rhodanate	0·5	0·7	0·4	5	334
112/35	„ „	0·1	91·5	4·8	6	343
„	„ „	0·3	6·8	4·3	6	312
„	„ „	0·5	0·7	0·7	6	384
28/36	„ „	0·3	20·1	10·8	4	311
113/35	*n*-Dodecyl thiocyanate	0·1	71·4	15·4	5	262
„	„ „	0·3	12·2	8·5	5	305
„	„ „	0·5	12·6	10·9	5	276
114/35	Cetyl thiocyanate	0·1	107·5	5·7	5	317
„	„ „	0·3	96·4	0·9	5	295
„	„ „	0·5	96·0	9·5	5	336
29/36	„ „	0·3	92·7	10·8	5	471
28/36	*secondary*-Alkyl thiocyanates C_{10}—C_{12}	0·3	101·0	5·7	4	297
26/36	*secondary*-Hexyl thiocyanate	0·3	99·9	2·5	5	434
„	„ Heptyl „	0·3	100·2	3·7	5	463
27/36	„ Octyl „	0·3	87·7	7·1	5	280
„	„ Nonyl „	0·3	85·7	9·0	5	424
26/36	„ Decyl „	0·3	100·8	2·6	5	412
27/36	„ Undecyl „	0·3	97·1	4·9	5	394
28/36	„ Dodecyl „	0·3	102·9	2·4	4	282
29/36	*β*-Butoxy-*β′*-thiocyanodiethyl ether ..	0·3	30·6	8·3	5	444
„	*tertiary*-Butyl thiocyanate	0·3	89·9	10·6	5	484
30/36	Methylallyl „	0·3	91·8	5·4	5	393

TABLE IV.

Toxicity of Naphthalene, some Derivatives, etc., to O. antiqua *eggs.*

Test No.	Material tested.	Conc. %	Hatch % Controls = 100	S.E.	n	Total eggs.
57/35	Naphthalene (in 20% benzene) ..	5·0	79·9	7·2	4	329
85/35	Tetralin	2·5	67·5	18·5	4	250
„	„	5·0	23·0	11·1	4	248
73/35	„	10·0	0	—	5	242
30/34	Decalin	10·0	90·3	8·2	6	404
73/35	„	10·0	80·0	5·4	5	234
107/35	α-Monochlornaphthalene	1·0	87·9	4·3	6	298
84/35	„	2·5	37·2	22·9	5	373
„	„	5·0	10·7	10·2	5	356
„	„ (in 20% benzene)	5·0	18·5	14·3	5	387
32/34	„ (in 10% decalin)	10·0	0	—	5	484
64/35	„ („ „)	10·0	6·1	2·7	6	561
„	„ (in 1·1% decalin) /casein-Turkey Red oil emulsion	10·0	2·7	1·7	6	393
85/35	Chlorinated naphthalene R77 (in 20% benzene)	10·0	7·2	3·7	4	236
86/35	Seekay Wax R93 (in 20% benzene)	10·0	28·7	12·1	5	282
„	„ R113 („ „)	10·0	71·1	6·3	5	358
„	„ R123 („ „)	10·0	68·2	8·3	5	277
30/34	„ R68 (in 10% decalin)	10·0	52·4	17·0	6	417
„	„ R93 („ „)	10·0	76·5	9·9	6	396
64/35	„ R68 („ „) /casein-Turkey Red oil emulsion	10·0	52·1	9·7	6	426
57/35	Benzene	20·0	105·8	5·3	4	354
„	Anthracene* (in 20% benzene) ..	5·0	55·2	18·2	4	325
56/35	Thionaphthene (in 10% benzene) ..	2·0	101·4	15·6	5	486

* Not a very satisfactory emulsion.

TABLE V.

Toxicity of Organic Bases to O. antiqua *eggs.*

Test No.	Material tested.	Conc. %	Hatch % Controls = 100	S.E.	n	Total eggs.
99/35	Nicotine	0·05	41·6	4·6	4	344
„	„	0·1	15·4	14·9	4	269
65/35	„	0·2	0	—	7	491
„	Anabasine	0·2	90·1	3·6	7	525
„	Pyridine	0·2	90·9	8·7	7	508
105/35	Quinoline	0·25	83·7	12·1	6	306
54/35	„	0·5	3·6	2·3	6	318
105/35	2-Methylquinoline (Quinaldine) ..	0·25	64·3	14·1	6	367
54/35	„ „	0·5	14·5	6·2	6	372
107/35	6-Methylquinoline	0·25	47·7	13·3	6	267
54/35	„	0·5	1·5	1·5	6	388
108/35	2:6-Dimethylquinoline (in 10% benzene)	0·25	92·9	8·1	5	173
55/35	„ („ „)	0·5	10·9	6·9	6	252
108/35	*iso*-Quinoline	0·25	5·0	2·7	5	225
55/35	„	0·5	1·2	1·2	6	263
„	Pyrrole	0·5	97·0	7·1	6	271
56/35	Carbazole (in 20% benzene)	0·5	68·3	5·5	5	382

Green Apple Aphis.

In the tables (VI-IX) of results of tests on eggs of *A. pomi* the per cent. hatch and its standard error (columns 4 and 5) have, except where otherwise stated, been derived from five replications. The results in the /35 tests were somewhat unsatisfactory and little reliance can be placed on any but very large differences. Greater experience and improved methods of handling the material made the results in the /36 and /37 series much more reliable.

TABLE VI.

Toxicity of Dinitrophenols and their Salts to A. pomi *eggs.*

Test No.	Material tested.	Conc. %	Hatch %	S.E.	Total eggs.
76/35	Sodium 2:4-dinitrophenate	0·25	**0·3**	0·3	1287
"	" "	0·5	**0**	—	1115
"	" 3:5-dinitro-*o*-cresylate	0·25	**0**	—	968
"	" "	0·5	**0**	—	1127
62/35	Controls (sodium oleate)	1·0	**23·4**	5·9	1175
"	Controls (untreated)	—	**34·6**	2·8	761
16/37	3:5-Dinitro-*o*-cresol (in 5% petroleum oil A)/sulphite-lye emulsion	0·05	**0**	—	2389
"	" " "	0·1	**0**	—	2250
"	3:5-Dinitro-*o*-cresol (in 1% petroleum oil B)/sulphite-lye emulsion	0·05	**0**	—	2095
"	" " " ..	0·1	**0**	—	2464
"	Petroleum oil A/sulphite-lye emulsion ..	1·0	**75·1**	1·7	1510
"	" " " ..	5·0	**74·0**	1·5	3043
"	Petroleum oil B/sulphite-lye emulsion ..	1·0	**65·7**	4·5	3069
"	" " " ..	5·0	**72·8**	4·5	2011
"	Controls (untreated)	—	**93·8**	2·3	1972

TABLE VII.

Toxicity of Thiocyanates to A. pomi *eggs.*

Test No.	Material tested.	Conc. %	Hatch %	S.E.	Total eggs.
83/35	*n*-Dodecyl thiocyanate	0·25	**1·3**	1·0	1245
"	Lauryl rhodanate	0·25	**2·3**	2·3	1354
"	Controls (untreated)	—	**51·3**	8·7	1782
7/36	*secondary*-Hexyl thiocyanate	0·25	**59·4**	3·4	766
"	" Heptyl "	0·25	**59·9**	2·2	1520
"	" Octyl "	0·25	**52·1**	1·3	1442
"	" Nonyl "	0·25	**59·7**	1·1	1838
"	" Decyl "	0·25	**29·9**	1·5	1778
"	" Undecyl "	0·25	**48·7**	1·7	712
"	" Dodecyl "	0·25	**39·8**	1·0	895
"	" Alkyl thiocyanates C_{10}—C_{12} ..	0·25	**73·1**	2·2	1415
"	Lauryl rhodanate	0·25	**0**	—	1080
"	β-Butoxy-β′-thiocyanodiethyl ether ..	0·25	**0**	—	1232
"	*tertiary*-Butyl thiocyanate	0·25	**70·7**	1·6	2087
"	Methylallyl thiocyanate	0·25	**38·9**	5·0	1391
"	Controls (untreated)	—	**93·0**	1·1	1437

TABLE VIII.

Toxicity of Naphthalene, some Derivatives, etc., to A. pomi *eggs.*

Test No.	Material tested.	Conc. %	Hatch %	S.E.	Total eggs.
76/35	Naphthalene (in 10% benzene)	2·0	22·7	2·8	601
80/35	Tetralin	2·0	24·6	4·3	784
"	Decalin	2·0	42·7	2·0	1673
"	α-Monochlornaphthalene (in 2% tetralin) ..	0·5	29·3	7·4	518
"	" (" ") ..	1·0	25·9	0·9	655
"	" (" ") ..	2·0	34·1	7·4	739
"	Chlorinated naphthalene R77 (in 2% tetralin) ..	2·0	4·8	1·7	1721
"	Seekay Wax R93 (in 2% tetralin) ..	2·0	23·9	6·4	650
"	" " R113 (" ") ..	2·0	17·0	6·4	1045
"	" " R123 (" ").. ..	2·0	14·5	2·6	767
76/35	Benzene	10·0	24·5	3·4	992
"	Anthracene (in 10% benzene)	2·0	0	—	958
80/35	Controls (untreated)	—	50·2	3·3	636
8/36	Naphthalene (in 10% benzene)	2·0	78·7	2·6	1543
"	Benzene	10·0	52·0	0·4	1659
"	Anthracene (in 5% benzene)	1·0	73·7	1·2	1433
"	,, (in 10% benzene)	2·0	58·1	4·9	1838
"	Controls (sodium oleate)	1·0	88·4	3·8	1443
"	Controls (untreated)	—	94·2	0·5	1646

TABLE IX.

Toxicity of Organic Bases to A. pomi *eggs.*

Test No.	Materials tested.	Conc. %	Hatch %	S.E.	Total eggs.
83/35	Nicotine	0·1	0·8	0·4	1655
"	Anabasine	0·1	44·6	6·0	1391
"	Pyridine	0·1	43·6	12·9	395
"	Quinoline	0·1	42·6	9·2	769
"	"	0·2	71·7	6·6	1879
"	Pyrrole	0·1	56·1	9·6	568
"	"	0·2	52·4	9·4	548
"	Controls (untreated)	—	51·3	8·7	1782
8/36	Nicotine	0·04	32·8*	1·6	746
"	"	0·07	20·4	0·6	1184
"	"	0·1	0	—	1064
"	Controls (sodium oleate)	1·0	88·4	3·8	1443
"	Controls (untreated)	—	94·2	0·5	1646
16/37	Nicotine	0·04	6·8	1·7	2357
"	"	0·06	8·8	2·4	2038
"	"	0·08	1·1	0·2	2211
"	Nicotine (in 5% petroleum oil A)/sulphite-lye emulsion	0·04	70·0	0·9	2079
"	" " " ..	0·06	33·9	3·4	1912
"	" " " ..	0·08	20·8	2·8	2251
"	Petroleum oil A/sulphite-lye emulsion ..	5·0	74·0	1·5	3043
"	Controls (untreated)	—	94·4	1·8	1984

* Mean of four replicates.

Table X.

Toxicity to O. brumata *eggs.*

Test No.	Material tested.	Conc. %	Hatch %	S.E.	*n*	Total eggs.
	Salts of Dinitrophenols.					
18/36	Sodium 2:4-dinitrophenate	0·25	80·0	—	3	10
"	" "	0·5	0	—	5	77
1/37	" "	0·15	62·1	4·4	6	241
"	" "	0·2	46·9	12·0	5	201
"	" "	0·25	56·9	8·1	5	242
18/36	Sodium 3:5-dinitro-*o*-cresylate	0·25	2·6	—	4	38
"	" "	0·5	0	—	4	34
1/37	" "	0·1	54·8	9·0	5	402
"	" "	0·15	41·2	9·7	5	486
"	" "	0·2	45·7	3·0	4	240
"	" "	0·25	31·9	5·0	5	184
	Thiocyanates.					
16/36	*secondary*-Hexyl thiocyanate	0·4	81·1	—	5	190
"	" Heptyl "	0·4	76·4	—	5	216
"	" Octyl "	0·4	86·7	—	5	180
"	" Nonyl "	0·4	87·4	—	5	87
"	" Decyl "	0·4	93·9	—	5	212
"	" Undecyl "	0·4	97·3	—	5	75
"	" Dodecyl "	0·4	97·6	—	5	81
1/37	*n*-Dodecyl thiocyanate	0·2	76·8	5·7	6	204
"	" "	0·4	53·6	9·1	4	440
17/36	*secondary*-Alkyl thiocyanates C_{10}—C_{12} ..	0·4	95·7	—	5	141
"	Lauryl rhodanate	0·2	97·8	—	5	90
"	" "	0·4	61·4	—	5	140
"	β-Butoxy-β′-thiocyanodiethyl ether ..	0·2	80·6	—	5	31
"	" " " ..	0·4	11·9	—	5	59
1/37	" " " ..	0·2	82·7	6·6	6	234
"	" " " ..	0·4	75·2	5·8	6	167
17/36	*tertiary*-Butyl-thiocyanate	0·4	100·0	—	5	88
"	Methylallyl "	0·2	92·4	—	5	145
"	" "	0·4	78·6	—	5	103
	Naphthalene, etc.					
18/36	Naphthalene (in 20% benzene)	5·0	93·7	—	5	63
"	Benzene	20·0	98·1	—	5	54
"	Anthracene* (in 20% benzene)	5·0	94·5	—	5	91
2/37	Diphenyl oxide	0·5	72·7	4·7	5	377
"	Diphenylene oxide	0·5	65·6	8·5	5	216
	Organic Base.					
18/36	Quinoline	0·3	76·4	—	4	55
"	"	0·5	47·8	—	5	23
	Controls.					
14/36	Controls (sodium oleate)	1·0	84·5	3·6	5	513
"	Controls (untreated)	—	92·8	1·4	5	367
16/36	" "	—	96·8	—	5	94
18/36	" "	—	96·1	—	5	52
1/37	" "	—	91·9	1·5	5	394
2/37	" "	—	90·3	1·9	5	223

* Not a very satisfactory emulsion.

Winter Moth.

Normally five lengths of apple shoot bearing Winter Moth eggs were used for a test of any one substance. Occasionally shoots proved to bear too few eggs and were ultimately discarded. In Table X, therefore, the number of replications used (column 6) to obtain the estimate of the mean per cent. hatch (column 4) and its standard error (column 5) is indicated. In some cases very few eggs indeed were recorded and the results of such tests are obviously less reliable. Standard errors are not given wherever their calculation appeared useless. For convenience, the results of all the control series have been collected together at the end of the table. They can be identified by means of their index numbers.

Fruit-tree Red Spider.

The results of two series of tests on Red Spider eggs are given. The first (Table XI) was done on eggs borne on raspberry canes. The numbers of hatched and unhatched eggs were recorded by Method 1 (29), in which counting

TABLE XI.

Toxicity to O. ulmi *eggs.*

Test No.	Material tested.	Conc. %	Hatch %	Total eggs.
22/34	Sodium 2:4-dinitrophenate	1·0	**33·7**	941
„	Sodium 3:5-dinitro-*o*-cresylate	1·0	**23·2**	647
„	Decalin/goulac emulsion	20·0	**64·5**	147
„	a-Monochlornaphthalene (in 5% decalin)/goulac emulsion	20·0	**36·3**	987
„	Seekay Wax R68 (in 20% decalin)/goulac emulsion	20·0	**15·8**	349
„	„ „ R68 (in 5% soap)	20·0	**16·6**	658
„	„ „ R93 („ „)	20·0	**12·8**	798
„	Controls (untreated)	—	**59·2**	484

is confined to random sample fields of view under a binocular microscope. Previous years' shells formed a large and variable proportion of the total and it was impossible to differentiate between them and newly hatched eggs. Furthermore, counts made before hatching began revealed such irregularity in the numbers of old shells (15-40 per cent.) that these could not be allowed for by any reasonable adjustment of the per cent. hatch. It was clear, however, that where a hatch of about 20 per cent., or less, was recorded, the kill could be regarded as almost complete. For example, counts made in another test revealed apparent hatches of 19·8 and 21·6 per cent. where a complete kill had obviously been obtained with a petroleum oil at concentrations of 10 per cent. and 6 per cent. respectively.

The second series (Table XII) was done on eggs laid on apple shoots. These were treated on March 16th, 1938, and subsequently dealt with according to Method 2 (29). The mean per cent. hatch and its standard error were derived from five replicates.

TABLE XII.

Toxicity to O. ulmi *eggs.*

Test No.	Material tested.	Conc. %	Hatch %	S.E.	Total eggs.
21/37	3:5-Dinitro-*o*-cresol (in 1% petroleum oil B)/ sulphite-lye emulsion	0·1	**0·9**	0·4	1317
„	β-Butoxy-β′-thiocyanodiethyl ether (in 1% petroleum oil B)/sulphite-lye emulsion ..	0·1	**29·9**	0·6	1588
„	Petroleum oil B/sulphite-lye emulsion ..	2·0	**60·0**	4·0	1324
„	Controls (untreated)	—	**86·0**	2·6	1648

Apple Sucker.

Some tests were made in 1937-38 on Apple Sucker eggs laid on apple twigs, like those made on Winter Moth eggs. It was found difficult to distinguish old shells from newly hatched eggs and there was also a tendency after certain treatments (e.g. the dinitrophenols) for nymphs to hatch out, only to die almost immediately. The results must, therefore, be accepted with reserve, although the variability of the counts appeared consistent with the errors of random sampling.

While the petroleum oil A at 5 per cent. (in sulphite-lye emulsion) was non-toxic, 3:5-dinitro-*o*-cresol in this medium gave an almost complete kill at 0·1 per cent. and was still highly toxic at 0·05 per cent. Nicotine at 0·08 per cent. in soap showed definite toxicity, whereas soap alone was non-toxic.

FIELD TRIALS.

MATERIALS AND METHODS.

Chemical.

The following materials were used in 1937:—

a. A stock emulsion consisting of 5 per cent. lauryl rhodanate (technical dodecyl thiocyanate) in 60 per cent. Grade E (20) petroleum oil emulsified by 10 per cent. Agral SR. This was used at 6 per cent. so as to give 0·3 per cent. of thiocyanate in 3·6 per cent. petroleum oil.

b. A wash containing the same concentrations of thiocyanate (0·3 per cent.) and oil (3·6 per cent.), but prepared by pouring into water a mixture of these ingredients with 0·5 per cent. of the emulsifier Whitcol J (27).

c. A wash similar to *b* but in which 0·3 per cent. mixed *secondary*-alkyl thiocyanates C_{10}-C_{12} was substituted for lauryl rhodanate.

d. A solution of 0·05 per cent. nicotine in 1 per cent. soap.

Preparation *a* was a highly stable emulsion, whereas *b* and *c* were of the quick-breaking type.

In 1938, concentrations were modified, the range of materials was extended and different emulsions were used. Lauryl rhodanate, a 75 per cent. preparation of β-butoxy-β'-thiocyanodiethyl ether, 3:5-dinitro-*o*-cresol and nicotine, each at 0·1 per cent., were applied in 5 per cent. emulsions of petroleum oil A (Grade E). With the exception of nicotine, which was added to the diluted emulsion, each of these was mixed with the requisite amount of oil before emulsification. In a further series of trials dinitrocresol was used in a 2 per cent. emulsion of petroleum oil B (Edeleanu extract) and nicotine was used at 0·1 per cent. and 0·05 per cent. in a 3 per cent. emulsion of petroleum oil A and at 0·05 per cent. in 0·5 per cent. soap solution.

The stock emulsions were prepared by milling the oil with one tenth its volume of sulphite-lye (60° Tw.) dissolved in sufficient water to make the final concentration of oil A 75 per cent. and of oil B 60 per cent. In some of the mixtures a water in oil emulsion was first obtained, but it was never difficult to secure inversion to the correct phase. Excellent emulsions were obtained and only those containing 60 per cent. of oil showed any tendency to cream.

Biological.

The trials were carried out on a plantation of Czar plums infested with eggs of both Leaf-Curling Plum Aphis (*Anuraphis padi* L.) and Hop-Damson Aphis (*Phorodon humuli* Schr.).

In 1937 spraying was done on February 11th. A single row of trees was divided into two blocks, each of six plots of three trees, and each spray was applied with a "Cascade" hand-pump machine to one plot in each block. None of the sprays caused any injury to the trees.

On May 13th the extent of the attack on each tree was estimated. Figures 1 to 5 were used to represent fifths of the tree affected by leaf-curl; thus a tree recorded as 1 showed leaf-curl on not more than about one-fifth of its branches. Similarly, intensity of attack was estimated as 1 slight, 2 moderate, or 3 severe. The product of the two figures gives a fair estimate of the Aphis damage. In assessing the results of this trial no clear distinction can be drawn between the effect of the sprays on the eggs and on the Aphides themselves, since several Aphides were observed hatching as early as February 10th.

In 1938 most of the sprays were applied on January 27th. Unsuitable weather delayed the application of the remainder till February 2nd, but all

treatments were completed long before hatching began. The earliest Aphides were not observed before February 18th.

The sprays were applied in the same way as in the previous year, but an improved lay-out was adopted. The trees in each of the 1938 trials were divided into four blocks, each containing one tree under each treatment. Aphis damage was estimated on May 25th in the same way as in 1937.

RESULTS.

1937.

The mean infestation per tree per plot is shown for each treatment in Table XIII.

TABLE XIII.

Field Trial on Plums, 1937.

Treatment.	Aphis attack.
A. 0·3% Lauryl rhodanate in 3·6% petroleum oil A/Agral SR emulsion ..	0·00
B. 0·3% „ „ 3·6% „ „ /Whitcol J emulsion ..	1·00
C. 0·3% C_{10}—C_{12} *sec.*-thiocyanates in 3·6% petroleum oil A/Whitcol J emulsion	5·00
D. 3·6% petroleum oil A/Whitcol J emulsion	5·33
E. 0·05% Nicotine in 1% soap (sodium oleate)	0·67
F. Controls (unsprayed)	14·50

	General Mean.	S.E.	S.E. Diff.	Sig. Diff. (P=·05).
Including controls ..	4·42	1·78	2·51	6·46
Excluding controls ..	2·40	1·85	2·62	7·27

An analysis of variance failed to reveal significant differences between spray treatments. Doubtless this was partly due to the inadequacy of the lay-out. Several of the trees in C and D were fairly heavily infested with Aphis and there can be no doubt that lauryl rhodanate was superior to the *secondary* thiocyanates. Where Whitcol J was used as emulsifier, the emulsions as first formed were poor, but they improved as spraying and agitation continued. The effect of this was seen in the records of C and D, for in each of these treatments Aphis attack was more severe in the first of the two plots sprayed. No such difference could be detected in B, where the high toxicity of the *normal* thiocyanate probably compensated any slight shortcoming in emulsification. Nicotine in soap appeared to be as highly toxic as lauryl rhodanate.

For the reason already stated, it is not clear from this trial how far the toxic effect of any of the above substances was due to ovicidal action alone.

A qualitative comparison of the effects of the various sprays on the eggs of the Mealy Aphis (*Hyalopterus arundinis* F.) was made on May 18th. Mealy Aphis was prevalent on the unsprayed plots, less so but irregularly distributed on the plots sprayed with oil emulsion alone, scanty where either nicotine or the *secondary* thiocyanate had been used, and absent from any of the plots sprayed with either of the emulsions containing the *normal* thiocyanate.

1938.

The treatments and mean infestations per tree are shown in Tables XIV and XV.

TABLE XIV.

Field Trial A on Plums, 1938.

Treatment.	Aphis attack.
0·1% 3:5-Dinitro-*o*-cresol in 5% petroleum oil A/sulphite-lye emulsion ..	1·00
0·1% β-Butoxy-β′-thiocyanodiethyl ether „ „ „ ..	0·75
0·1% Lauryl rhodanate „ „ „ ..	0·50
0·1% Nicotine .. .. „ „ „ ..	1·00
5% Petroleum oil A/sulphite-lye emulsion	0·75
2% Petroleum oil B/ „ „	0·50
Controls (unsprayed)	8·25

General Mean.	S.E.	S.E. Diff.	Sig. Diff. (P=·05).
0·69	0·46	0·65	1·34

TABLE XV.

Field Trial B on Plums, 1938.

Treatment.	Aphis attack.
0·05% Nicotine in 3% petroleum oil A/sulphite-lye emulsion	0·50
0·1% „ „ „ „ „	0·75
0·05% „ 1% soap (sodium oleate)	4·00
0·1% 3:5-Dinitro-*o*-cresol in 2% petroleum oil B/sulphite-lye emulsion ..	0·50
Controls (unsprayed)	9·75

General Mean.	S.E.	S.E. Diff.	Sig. Diff. (P=·05).
1·44	0·71	1·00	2·26

Since each treatment reduced the attack, the data from the unsprayed plots were excluded from the analyses of variance that have provided the statistical information below each Table. On the other hand, certain treatments were

included in the analyses but are omitted here as irrelevant to the present discussion.

Nicotine in soap greatly reduced Aphis attack, but was not as effective as the other sprays tested. Its toxicity was evidently due to its ovicidal properties since it was applied to eggs only and not, as in the previous season's trial where its toxicity was greater, to hatching Aphides as well as to unhatched eggs. The possible toxic effect of the soap itself was also investigated. Three trees sprayed with soap alone showed a mean infestation per tree of 7·0. As these were at the less heavily attacked end of the plantation, the infestation cannot be said to differ from that of unsprayed trees.

The comparative efficiency of dinitrocresol, β-butoxy-β'-thiocyanodiethyl ether, lauryl rhodanate and nicotine is not apparent, owing to the unexpectedly high toxicity of the 5 per cent. petroleum emulsion in which these substances were incorporated.

0·1 per cent. of 3:5-dinitro-*o*-cresol requires not less than 5 per cent. of petroleum oil A for its solution, consequently less than that amount of oil could not well have been used. Although some kill was expected, it was not anticipated that the toxicity of the oil would be so high as to mask the effect of the other materials. In the previous season's trial, 3·6 per cent. of petroleum gave a good kill but, at that time, it appeared likely that this toxicity was largely due to action on hatched and hatching Aphides. The apparent superiority of the nicotine-oil emulsion over nicotine-soap may be due solely to the toxicity of the 3 per cent. of petroleum oil.

The eggs of the plum Aphides are evidently very easily killed, a conclusion amply borne out in various trials of low concentrations of tar oils carried out at the same time as the trials reported here.

DISCUSSION.

DINITROPHENOLS AND THEIR SALTS.

Without exception 3:5-dinitro-*o*-cresol, whether as the free phenol or as its sodium salt, proved to be more toxic than 2:4-dinitrophenol, thus confirming the results of Tattersfield *et al.* (30). 13.364

In laboratory tests against *A. pomi* eggs, concentrations as low as 0·05 per cent. were completely toxic. Against Red Spider eggs, too, the material showed great promise, being very toxic at 0·1 per cent. It was less effective against Lepidoptera and was rather less toxic to Vapourer than to Winter Moth eggs. Even so it compared very favourably with the most toxic thiocyanates.

From eggs of Vapourer Moth and Apple Sucker, treated with dinitrocresol, the insects were frequently able to hatch but they died immediately. This tendency has been noticed by Gimingham and Tattersfield (9).

THIOCYANATES.

The thiocyanates showed a very wide range of ovicidal power but only *n*-dodecyl thiocyanate and β-butoxy-β'-thiocyanodiethyl ether showed a promising degree of toxicity. From the point of view of the relation between chemical constitution and toxic action, the most striking effect was the reduction (almost elimination) of toxicity in passing from the *primary* to the *secondary* series. It is clear, too, that cetyl thiocyanate compared very unfavourably with the shorter chain compound, dodecyl thiocyanate. Both these conclusions agree with previous observations on the efficiency of these materials as contact insecticides. Bousquet *et al.* (1) showed that toxicity to various insects reaches a peak about the C_{12} compound, and they mention also the comparative lack of promise shown by the secondary thiocyanates. Kearns and Martin (15) found cetyl thiocyanate less ovicidal than dodecyl thiocyanate to *A. pomi* eggs.

Even more toxic, on the whole, than dodecyl thiocyanate was β-butoxy-β'-thiocyanodiethyl ether, although the concentrate contained only 75 per cent. of active material.

In the laboratory the two most toxic thiocyanates killed about 80 per cent. of Vapourer Moth eggs at concentrations of the order of 0·3 to 0·5 per cent. but they were only moderately toxic to Winter Moth eggs at 0·4 per cent. 0·25 per cent. was sufficient to give a complete kill of eggs of *A. pomi*.

NAPHTHALENE, DERIVATIVES, ETC.

The introduction of one chlorine atom into the α-position of the naphthalene molecule greatly increased toxicity to Vapourer Moth eggs, but this effect progressively diminished with the introduction of further chlorine atoms. Reduction of naphthalene to tetralin increased toxicity, but further reduction reversed the effect, decalin being no more toxic than naphthalene. This result of hydrogenation agrees with Tattersfield's observation (30) on eggs of *S. tetralunaria*.

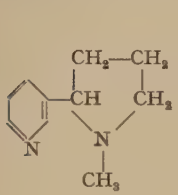
Against *A. pomi* eggs there is, apparently, no correlation among the chlorinated naphthalenes between chlorine content and toxicity. Only one of them, R77, was significantly more toxic than naphthalene itself. Tetralin, also, was no more toxic than naphthalene, whilst decalin was virtually non-toxic.

Against Red Spider eggs, the toxicity of the chlorinated naphthalenes appears to increase with increasing chlorine content.

ORGANIC BASES.

Nicotine (I) was much the most toxic of the bases tested and contrasts strongly with the closely related anabasine (II) and pyridine (III) with which it was compared against Winter Moth and *A. pomi* eggs. Whether this toxicity

is a property of the methyl-pyrrolidine ring (IV) as compared with the piperidine ring (V) is a matter for further investigation.



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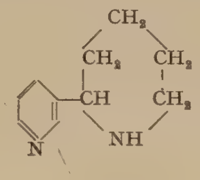
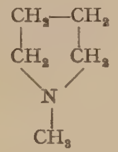


II

III



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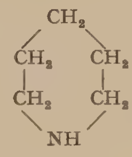

V

The effect of nicotine on Winter Moth eggs has not yet been adequately investigated but they appear to be less susceptible than the eggs of the Vapourer Moth.

Of the remaining bases, all the quinoline derivatives were highly toxic to eggs of the Vapourer Moth at 0·5 per cent. and one, *iso*-quinoline, was very toxic at 0·25 per cent. They can probably be placed in the following order of toxicity to these eggs:—

iso-quinoline>6-methylquinoline>quinoline>2 : 6-dimethylquinoline = 2-methylquinoline.

Pyrrole and carbazole were non-toxic at 0·5 per cent.

To *A. pomi* eggs, quinoline and pyrrole were non-toxic at 0·2 per cent. and to Winter Moth eggs, quinoline was not very toxic even at 0·5 per cent.

Of all the bases examined, nicotine received particular attention because of interest in the nature of its ovicidal action, the physiological mechanism of which is by no means fully understood. It was repeatedly observed that eggs treated with nicotine appeared to develop normally until hatching time, when the embryo either failed to hatch or perished in the attempt. A similar aborting action of nicotine has been reported by Marchal (19), Feytaud (6, 7) and Misaka (21).

Three explanations that can be advanced are:—

(1) that nicotine forms some chemical or physical complex toxic to the insect when it subsequently attempts to hatch out,

(2) that such a complex prevents hatching by interfering with respiration or by other mechanical means such as might result from a hardening of the chorion,

(3) that nicotine penetrates the egg and affects metabolism in such a way that the insect becomes incapable of hatching, even though interference with the development of the embryo may not be obvious.

1. *Delayed toxicity*. The first hypothesis has been favoured by Feytaud (7) who concluded that nicotine applied to the eggs of *Polychrosis botrana* poisoned the larvae when eventually they attempted to eat their way out.

Kearns *et al.* (14) considered that nicotine does not behave as a simple ovicide to the Apple Sawfly but persists in the calyx tissue surrounding the eggs and kills the larvae when they eat it.

Although a stomach-poison action is intelligible for mandibulate insects, it is clearly inapplicable to the eggs of Aphides. Moreover, Table XVI shows that it does not fully explain results obtained with eggs of the Vapourer Moth—a mandibulate insect.

TABLE XVI.

Effect of Nicotine on O. antiqua *eggs.*

Test.	Treatment.	Percentage of eggs hatched.	part-hatched.	unhatched.
63/35	Nicotine 0·05%	25	32	43
	„ 0·01%	15	37	48
	Untreated	63	2	35
99/35	Nicotine 0·2%	0	55	45
	Untreated	75	2	23

In this Table part-hatched eggs are those containing fully formed but dead larvae that had made some evident attempt to hatch out. In each of them the larva had eaten a portion of the shell but had died without hatching. In some, the hole in the chorion was no bigger than a pinprick but in others was as large as in a hatched egg, and the dead larva could be seen either within the egg or partly protruding from it.

The unhatched eggs bore no signs of attempted hatching. Some would have failed to hatch anyway, but the remainder, represented by the difference in the percentage unhatched between treated and untreated sets of eggs, cannot have died as a result of the larvae eating poisoned egg shells but must have succumbed to some other insecticidal action. In test 63/35, where the data are derived from four egg-batches only and where the natural mortality is somewhat high, the percentage of unhatched eggs does not, in either of the treated series, differ significantly from that in the untreated (S.E. Diff. = 5·6, Sig. Diff. = 14, P = ·05). In test 99/35, however, where seven egg-batches are available and the natural mortality is lower, the difference is highly significant (S.E. Diff. = 4·5, t = 4·88, P = < ·01).

2. *Mechanical effect.* There appears to be no good reason to suppose that nicotine forms a film over the egg or its micropyle sufficient to interfere with respiration and so cause death by stifling. Such a physical action has been suggested by Staniland *et al.* (28) to account for the toxicity of petroleum oils to certain types of egg but, as an explanation of the ovicidal action of nicotine,

it has been rejected by Misaka (21). Possibly, however, some interaction between the insecticide and the constituents of the egg shell results in a hardening or toughening of the chorion. Although this idea of mechanical interference with hatching is compatible with the observation that treated eggs do not hatch despite the lack of apparent effect on embryological development, the evidence suggests rather that nicotine acts by penetration.

3. *Penetration.* Although Feytaud concluded (7) that the aborting effect of nicotine on eggs of *Polychrosis* is due simply to a delayed stomach-poison action, he had suggested in an earlier paper (6) that it is due to an internal insecticidal action on the developing embryo.

Misaka (21), working with *Chilo simplex*, found that although a high concentration (0·03 per cent.) of nicotine sulphate killed all the eggs, lower concentrations killed only those in which embryological development was well advanced. He concluded that nicotine vapour penetrates the shells and kills embryos that have fully formed nerves and tracheae but merely retards the development of younger embryos, whose nervous system is less well developed.

Table IX shows that nicotine was more toxic to *A. pomi* eggs in 1937-38 than in the preceding season. In the series showing the higher level of toxicity, nicotine was applied to the eggs about two weeks before hatching, whereas in the other series more than five weeks elapsed between treatment and hatching. It has been shown by Peterson (25, 26) and, less conclusively, by Lees (17, 18) that eggs of this aphis are killed by nicotine preparations more readily when the insecticide is applied towards hatching time.

Peterson suggested that the eggs become more susceptible after their outer glutinous covering splits, a few days before hatching. This splitting scarcely ever occurred in the present experiments. The egg almost invariably showed no sign of hatching till the chorion was ruptured by the Aphis's egg burster, when both chorion and outer covering split together.

That nicotine penetrates the egg shell and then interferes with the development of the embryo appears to the present authors to be the most likely explanation of its toxic action. This view is supported by the fact that nicotine was less toxic to *A. pomi* eggs when incorporated in a mayonnaise emulsion of petroleum oil than when used with sodium oleate (Table IX). It seems reasonable to suggest that the oil impeded penetration and that the soap facilitated it.

GENERAL DISCUSSION.

Of all the materials tested, most can be dismissed as showing little promise as winter ovicides. These include all but two of the thiocyanates, the naphthalene derivatives and allied substances, and the bases other than nicotine and some of the quinoline and *iso*-quinoline compounds. The quinoline

and *iso*-quinoline derivatives were not very toxic but may repay further investigation along the line of synthesizing a toxic molecule. The materials worthy of extended trial are 3:5-dinitro-*o*-cresol, *n*-dodecyl thiocyanate (lauryl rhodanate), β-butoxy-β'-thiocyanodiethyl ether and nicotine. All these are suitable for use as supplements for winter petroleum washes, and commercial preparations of some of them are now available.

SUMMARY.

The ovicidal properties of forty-four organic preparations, most of them chemically individual substances, have been investigated in the laboratory, using as test objects the eggs of the Vapourer Moth (*Orgyia antiqua* L.), the Green Apple Aphis (*Aphis pomi* De Geer), the Winter Moth (*Operophtera brumata* L.), the Fruit-tree Red Spider (*Oligonychus ulmi* C. L. Koch), and the Apple Sucker (*Psyllia mali* Schmidt). The preparations were mainly dinitrophenols, thiocyanates, naphthalene derivatives and organic bases. 3:5-Dinitro-*o*-cresol, *n*-dodecyl thiocyanate (lauryl rhodanate), β-butoxy-β'-thiocyanodiethyl ether and nicotine were the most toxic and the first named was outstanding. Wherever sufficient data were available, attempts have been made to correlate differences in toxicity with modifications of molecular structure.

The four materials mentioned were further tested in field trials on plum trees infested with Leaf-Curling Plum Aphis (*Anuraphis padi* L.) and Hop-Damson Aphis (*Phorodon humuli* Schr.), and largely bore out their laboratory promise.

The mechanism of the ovicidal action of nicotine is discussed in some detail.

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THE WATER CONDUCTIVITY OF THE GRAFT UNION IN APPLE TREES, WITH SPECIAL REFERENCE TO MALLING ROOTSTOCK No. IX

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INTRODUCTION.

Apples in this country are usually grafted on a rootstock which consists of a piece of stem together with its root system. Hence the observed effect of any rootstock on the scion may be due to the influence of the root system, to that of the piece of rootstock stem, to the nature of the union between rootstock and scion, or to a combination of any or all of these factors. The relative importance of the root system and the stem piece has already been investigated by others, both in single- and in double-worked trees (4, 5, 6, 7, 10, 11). The nature of the union, so far as fruit trees are concerned, has been studied chiefly where incompatibility, either partial or complete, exists between stock and scion (1).

The purpose of the present paper is to describe some experiments carried out to determine the effect of the union in grafted apple trees on the water conductivity of the piece of stem containing that union, and, in general, to compare the water conductivity of unions in which the dwarfing rootstock, Malling No. IX, forms one component with the conductivity of unions of which a vigorous stock (Malling No. XII or a seedling stock) was a component. The data which will be presented show that a union of M.IX with certain scion varieties or with stock M.XII constitutes a resistance to the flow of water in the stem, and that such unions, so far as water conductivity is concerned, appear to be less efficient than unions between a vigorous stock and a scion variety.

MATERIAL.

The first determinations were carried out on a few miscellaneous lots of material comprising:—

(1) maiden trees of Lane's Prince Albert on stocks M.IX and M.XII, from East Malling ;

(2) three-year-old trees of Lane's Prince Albert on stocks M.IX and on a free stock, from a commercial nursery ;

(3) two-year-old trees of Bramley's Seedling on stock M.IX and on Crab stock, from Long Ashton.

As will be seen later, these preliminary observations were too few in number to allow any definite conclusions to be drawn. Later, a series of double-worked trees, with Stirling Castle scions, used by Vyvyan (11) for an investigation into the relative importance of stock and intermediate piece of stem in double-worked trees, became available; and, as approximately eighty trees were available for experimental work, the conclusions drawn from the data collected appear to have considerable significance.

It was clear that simple determinations of the conductivity of a piece of stem containing the union would yield results of relatively little value, as in all probability the most important factor affecting the conductivity would be the size of the stem. This trouble could be obviated by expressing the conductivity on a unit cross-sectional area basis; but this is not always possible because a considerable bulge often occurs at the union, so that the mean cross-sectional area of a piece of stem 10 or 15 cm. long and containing a bulge is difficult to determine accurately. In order to obtain a clearer picture of the water relations of the trees it seemed desirable to make conductivity measurements for each tree on (1) a portion of rootstock stem immediately below the graft union, (2) a piece of stem containing the union and prepared so that as far as possible it contained equal lengths of the two components of the union, and (3) a portion of the scion stem immediately above the union. When this was done it was possible to see to what extent, if any, the union formed a barrier to the flow of water. With the double-worked trees a modification of this procedure, described below, had to be adopted.

PREPARATION OF MATERIAL AND EXPERIMENTAL METHODS.

(1) The trunks of the trees used for the preliminary observations were sawn into 10 cm. lengths and the ends of the stem pieces pared with a razor. To these a head of water equivalent to 16 cm. of mercury was applied and the amount of water transmitted through the piece of stem in ten minutes determined.

(2) The treatment of the double-worked trees was somewhat different. Here each tree consisted of three parts: rootstock, intermediate and scion. The stocks and intermediates were always either M.IX or M.XII, and all four possible combinations were used, so that four types of tree were available as shown below:—

Scion.	*Intermediate.*	*Stock.*
Stirling Castle	M.XII	M.XII
,, ,,	M.IX	M.XII
,, ,,	M.XII	M.IX
,, ,,	M.IX	M.IX

The trees had been worked in the winter of 1931-32, and their subsequent behaviour has been described by Vyvyan (11). Altogether eighty-four trees were available, and the conductivity of these was determined during the winter 1937-38. Of these trees, sixty-four had been grown in plots arranged in complete Latin squares and the remainder in broken Latin squares. In the conductivity measurements it has not been possible to make full use of the statistical lay-out of the original experimental plots in which the trees had been grown. Owing to occasional accidents, in no case could a complete set of data be obtained for the trees of any one Latin square. Hence all the data for each type of tree have been treated as though the trees were all members of a single population, mean values being determined and the standard errors of the means calculated.

Ideally, measurements of conductivity should have been made for each tree on stock, lower union, intermediate, upper union and scion. Owing to the short length of the intermediate pieces this was not possible. The pieces of stem containing the unions had to be 15 cm. long, and this left no intermediate available for experimental work. Owing to the pronounced bulge which often occurred when M.IX was one of the components of the union, pieces shorter than 15 cm. could not be fixed satisfactorily to the apparatus. To overcome this difficulty half the number of trees of each type were cut to give stock, lower union, upper union, and scion and the other half to give stock, intermediate, and scion. Even so, the pieces of intermediate used were only 7·5 cm. long, and to make the data collected from these pieces comparable with the conductivity measurements on the 15 cm. pieces, the observed conductivity was halved. That this procedure was admissible seemed likely (2, 3), but the assumption on which it was based was tested experimentally, the relevant data being given below. The conductivity of 15 cm. pieces of scion was determined by the method described below and then a length of 3·75 cm. was removed from each end of these pieces and the conductivity of the remaining 7·5 cm. determined. The data are incorporated in Table I.

TABLE I.

Conductivity of 15 cm. and 7·5 cm. pieces of stem, in gm. water in 15 mins. for pressure of 30 cm. of mercury.

Experiment.	15 cm. pieces.	7·5 cm. pieces.
1	18·0	34·3
2	7·5	17·0
3	14·5	26·0
4	11·5	25·0
5	14·0	29·5
6	15·6	27·2
Mean	13·5	26·5

These figures indicate that the procedure adopted was permissible. Even were some slight error introduced by this procedure it would not upset the main conclusions which have been drawn as they are based on a comparison between the various types of trees used, and all the trees were treated alike.

The actual conductivity measurements here were made using negative pressure. The pieces of stem were freed from air in the manner described in a previous paper (12). The apparatus used was similar to that employed previously (12), but it was modified so as to allow of up to six determinations being carried out simultaneously.

RESULTS.

(1) Maiden Trees of Lane's Prince Albert on Stocks M.IX and M.XII.

Table II.

Conductivity as gm. of water transmitted through 10 *cm. pieces of stem in* 10 *minutes with a pressure equivalent to* 16 *cm. of mercury.*

	Stem below union.	Stem containing union.	Stem above union.
Stock M. XII.			
Tree 1	10·86	9·12	11·88
2	9·30	7·67	6·17
3	6·40	7·42	9·63
Mean	8·85	8·07	9·23
Stock M. IX.			
Tree 1	4·20	1·82	3·68
2	1·60	1·12	2·65
3	3·18	1·64	2·35
Mean	2·99	1·53	2·79

Clearly, not only is the conductivity here much less on the smaller trees on stock M.IX, but the union does appear to offer a considerable resistance to the flow of water. This is well shown if the conductivity of the piece of stem containing the union is expressed as a percentage of the mean of the conductivities of the stock below and the scion above. The values obtained are 53·3% for stock M.IX and 90·6% for stock M.XII.

(2) Three-year-old Lane's Prince Albert Trees on Stock M.IX and on Free Stocks.

The trees on the free stock appeared quite healthy but were not vigorous, being no larger than the trees on M.IX.

TABLE III.

Conductivity as gm. of water transmitted through 10 *cm. pieces of stem in* 10 *minutes with a pressure equivalent to* 16 *cm. of mercury.*

	Stem below union.	Stem containing union.	Stem above union.
Free Stock.			
Tree 1	40·90	41·15	53·50
2	57·10	45·35	100·12
Mean	49·00	43·25	76·81
Stock M. IX.			
Tree 1	44·70	30·15	62·30
2	34·60	31·55	51·00
Mean	39·65	30·85	56·65

There is relatively little evidence here that the union with M.IX stock forms a barrier to the flow of water ; but if the conductivity of the union is expressed as a percentage of the mean of the conductivity of the stock and scion the values 65·4% for M.IX stock and 73·5% for the free stock are obtained.

(3) TWO-YEAR-OLD BRAMLEY'S SEEDLING ON STOCK M.IX AND ON CRAB STOCKS.

TABLE IV.

Conductivity as gm. of water transmitted through 10 *cm. pieces of stem in* 10 *minutes with a pressure equivalent to* 16 *cm. of mercury.*

	Stem below union.	Stem containing union.	Stem above union.
Crab Stock.			
Tree 1	16·00	13·18	43·12
2	24·70	18·98	43·22
Mean	20·35	16·08	43·17
Stock M. IX.			
Tree 1	13·20	6·28	21·77
2	9·33	5·20	14·03
3	9·15	9·21	32·52
Mean	10·56	6·90	22·77

Here the smaller trees on M.IX stock give lower values for the conductivities, but the effect of the union is not very marked. The conductivity of the union expressed as a percentage of the mean conductivities of the stock and scion is 50·2% for the Crab stock and 41·4% for stock M.IX.

(4) Double-worked Trees of Stirling Castle.

As mentioned earlier, trees of four types were available and for each combination of scion, intermediate and stock, conductivity determinations were made on scion, upper union, intermediate, lower union and stock. For reasons already outlined, all the determinations could not be made on every tree, but for each combination twenty or twenty-one determinations were made for the stock and scion, and ten or eleven for the upper union, intermediate and lower union.

(a) Diameters of Pieces Used.

The "average" diameter of each piece used was obtained by averaging the diameters at the upper and lower ends. With many unions a pronounced bulge occurred at the union, and the diameters given in Table V do not take this into account. The presence of this bulge makes any accurate estimate of the amount of conducting tissue at the union difficult, and makes it impossible to express accurately the conductivity measurements on a unit cross-sectional area basis.

Table V.

Diameters in cm. of stem pieces used. Mean values and standard errors.

Combinations.	Scion.	Upper union.	Inter-mediate.	Lower union.	Stock.
Stirling Castle /M. XII/M. XII	2·52 ±0·12	2·78 ±0·19	2·94 ±0·19	3·01 ±0·19	3·21 ±0·14
Stirling Castle /M. IX/M. XII	1·74 ±0·07	2·18 ±0·15	2·79 ±0·14	2·31 ±0·12	2·52 ±0·05
Stirling Castle /M. XII/M. IX	1·58 ±0·05	1·74 ±0·05	1·97 ±0·10	2·25 ±0·10	2·38 ±0·08
Stirling Castle /M. IX/M. IX ..	1·45 ±0·04	1·71 ±0·08	2·16 ±0·05	1·96 ±0·09	2·06 ±0·05

With each combination there is a regular decrease in diameter in passing from stock to scion with the exception of the trees Stirling Castle/M.IX/M.XII, where the intermediate is of greater diameter than either stock or scion and the trees Stirling Castle/M.IX/M.IX, where the value for intermediate is greater than that for the stock, but in this case the difference is not significant.

(b) Conductivities.

The conductivities of these stem pieces are given in Table VI.

A noticeable feature here is that while the diameter of the stem increases from above downwards the conductivity increases from below upwards. The intermediate always has a greater conductivity than the stock, and the scion, with one exception, has a greater conductivity than the intermediate. This agrees with the figures for the single-worked trees where the conductivity of the scion usually exceeded that of the stock.

TABLE VI.

Mean values and standard errors of gm. of water passed through a 15 cm. length of stem in 15 minutes with a pressure equivalent to 30 cm. of mercury.

Combinations.	Scion.	Upper union.	Inter-mediate.	Lower union.	Stock.
Stirling Castle /M. XII/M. XII	30·3 ± *1·66*	25·3 ± *2·06*	25·0 ± *2·44*	18·0 ± *1·57*	18·6 ± *2·64*
Stirling Castle /M. IX/M. XII	19·6 ± *1·80*	15·2 ± *2·10*	23·0 ± *1·52*	11·1 ± *2·63*	12·2 ± *1·24*
Stirling Castle /M. XII/M. IX ..	22·1 ± *1·52*	17·6 ± *2·02*	17·4 ± *1·56*	11·1 ± *1·29*	10·7 ± *0·70*
Stirling Castle /M. IX/M. IX ..	13·9 ± *0·86*	9·4 ± *0·78*	10·9 ± *1·11*	7·6 ± *0·98*	7·2 ± *0·64*

With the double-worked trees differences between the types of tree are of little interest, as the conductivity is manifestly influenced very largely by the size of the tree. This difficulty can to a great extent be obviated if the conductivities of the union, the intermediate, and the stock are expressed as a percentage of the conductivity of the scion of the same tree, as shown in Table VII.

TABLE VII.

Mean values and standard errors of conductivities of stem pieces expressed as percentage of scion conductivity.

Combinations.	Upper union.	Inter-mediate.	Lower union.	Stock.
Stirling Castle/M. XII/M. XII ..	89 ± *8·3*	80 ± *7·4*	63 ± *5·2*	62 ± *8·4*
Stirling Castle/M. IX/M. XII ..	94 ± *13·9*	110 ± *10·0*	62 ± *7·8*	68 ± *7·8*
Stirling Castle/M. XII/M. IX ..	78 ± *10·4*	90 ± *10·3*	45 ± *5·6*	52 ± *4·9*
Stirling Castle/M. IX/M. IX ..	74 ± *6·3*	80 ± *7·4*	63 ± *7·3*	54 ± *4·9*

Two points of interest emerge from this Table. Firstly, the conductivity of the intermediate is greatest in those combinations where intermediate and rootstock are unlike. Secondly, conductivity of stock, expressed as a percentage of that of the scion, is lowest in the trees in which M.IX is the stock.

TABLE VIII.

Conductivities of stem pieces with union as percentages of the mean of conductivities of stem above and below union.

Combinations.	Upper union.	Lower union.
Stirling Castle/M. XII/M. XII ..	91	83
Stirling Castle/M. IX/M. XII ..	70*	63*
Stirling Castle/M. XII/M. IX ..	89	79
Stirling Castle/M. IX/M. IX ..	77*	84

Finally, to obtain fuller information as to the efficiency of the graft union, the conductivities of the pieces of stem containing the unions have been expressed as percentages of the mean of the conductivities of the stem above and the stem below the union. The values obtained are given in Table VIII.

In the unions marked above with an asterisk a pronounced bulge occurred. In the lower union of the trees Stirling Castle/M.XII/M.IX no bulge occurred, but there was an over-growth at the base of the intermediate. Reference to this Table shows that all the unions of which M. IX forms one component have given low values.

DISCUSSION.

Discussion of the data presented will be confined almost entirely to the possible effect of the graft union in restricting the flow of water to the shoot system of the tree. It is not proposed to discuss the possible importance of the conductivity of either the stock itself or the intermediate. As far as the union itself is concerned it is clear that in none of the unions examined is there an almost complete barrier to water transport such as Chang (1) found in certain incompatible unions in pears, peaches and plums.

Reference, especially to the first Table containing conductivity data for the maiden trees of Lane's Prince Albert and to the final Table for the double-worked trees of Stirling Castle, shows, however, that unions in which M.IX is one component do appear to be less efficient for water conduction than unions in which M.IX is combined with itself, or M.XII is combined either with itself or with a scion.

It might be supposed that in a perfect union the conductivity of the piece of stem containing the union would exactly equal the mean of the conductivities of the two components. This is not so. When stock and scion are grafted together the cut surfaces soon develop callus in compatible unions (1), and water passing up the scion has to pass across this callus "bridge". Since this is composed of living cells, it probably has a high resistance to water flow. As soon as new xylem is formed, most, and later probably all, of the water transmitted to the scion is conducted past the union through the new tracheae. Just at the union the amount of conducting tissue is less than that in either stock or scion. There, only the new xylem is effective, while above and below the union all the xylem is available for water transport. Therefore, in unions in which little new xylem is produced, either because the stock is a dwarfing one or the components of the union are not compatible, the union might be expected to appear relatively inefficient. With increasing age the difference between the amount of conducting tissue at the union and in the stem above and below it becomes less. With the six-year-old double-worked trees, however, there is

evidence that the unions in which M.IX is one component are relatively somewhat inefficient. This inefficiency is really greater than is apparent. In the unions with M.IX, a bulge, often of considerable dimensions, develops, and the diameter of the stem at the union may be double that of the stock stem below or the scion stem above. The presence of such a bulge probably means a great increase in the amount of conducting tissue in the neighbourhood of the union, and this will tend to increase the conductivity. Undoubtedly, the bulging is accompanied by a marked distortion of the conducting elements, and this would tend to decrease the conductivity. The net result of these two opposing tendencies, and possibly of other factors the importance of which is not yet realized, is, as the data presented show, a reduced conductivity of the unions. Further work may reveal cases where the effect of the additional conducting tissue of the bulge at the union actually causes the conductivity at this point to exceed the conductivity of either stock or scion.

The data presented certainly indicate that the graft union introduces an additional resistance to the flow of water in the trees, and that this additional resistance is greater in those unions where stock M.IX is one of the components than in the other combinations studied. It is possible, however, that the use of short stem pieces for the conductivity measurements may have given a somewhat exaggerated picture of the effect of the union. If longer stem pieces had been used, the reduction in conductivity caused by the union would, in all probability, have been much less. This possibility, however, in no way affects the conclusion which has been drawn from the data, namely, that unions in which stock M.IX is one component present a greater resistance to water flow than the other unions studied, although it does render it more difficult to assess accurately the full significance of the data so far as the water economy of the intact tree is concerned. It may be pointed out, however, that certain growth characters of trees on M.IX stock, namely the presence of small leaves and short internodes, and the early cessation of seasonal shoot growth (8, 9), are characters which might be associated with a slight internal water deficit in the plant. Finally, there remains the possibility that the reduced conductivities of the unions between stocks M.IX and M.XII and between M.IX and certain scion varieties is an expression of partial incompatibility.

SUMMARY.

1. The water conductivity of the graft union in a number of apple trees, both single- and double-worked, has been measured.

2. The aim has been to compare the efficiency of unions in which Malling IX stock was one component with that of unions in which a vigorous stock was combined either with itself or with a scion.

3. The conductivity of the unions has been expressed as a percentage of the means of the conductivities of the stem above and the stem below the unions. It has been shown that unions of M.IX with certain scion varieties, and unions of M.IX with M.XII are less efficient than unions of M.XII with M.XII, M.IX with M.IX, M.XII with certain scion varieties, and seedling stocks with certain varieties.

4. When expressed as a percentage of the conductivity of the scion, the conductivity of the intermediate in double-worked trees was greatest when intermediate and stock were unlike.

5. When expressed as a percentage of the conductivity of the scion, the conductivity of the stock in the double-worked trees was less when M.IX was the stock than when M.XII was the stock.

6. The extent to which the union in these cases influences the water economy of the tree cannot fully be assessed, but it is pointed out that certain growth characters of trees on M.IX stock are features that might be expected if the water supply to the shoots was somewhat restricted.

7. It is not possible to say whether the reduced efficiency of unions in which stock M.IX forms one component is an expression of partial incompatibility between stock and scion.

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BOOK REVIEWS

STATISTICAL TABLES FOR BIOLOGICAL, AGRICULTURAL AND MEDICAL RESEARCH. By R. A. FISHER and F. YATES. (Oliver and Boyd, Edinburgh, 1938, pp. 90. 12s. 6d.)

In recent years there have appeared in this Journal and elsewhere many papers dealing with the statistical aspects of the many sided problems of agricultural and horticultural research. Some of these papers describe methods by which the research worker, having designed his experiments in accordance with certain fundamental principles, may himself analyse his numerical results. As new statistical methods have been evolved to meet the needs of increasingly complex research, statisticians have provided tables of various mathematical functions to assist in and simplify their use. Hitherto these tables have been scattered in various publications, sometimes not very accessible to those who would wish to use them. It was thus high time for a new book of statistical tables which would collect in one volume those now commonly recommended. Many of these have originated from Rothamsted and it is, therefore, specially suitable that Professor Fisher and Mr. Yates should now meet the demand which they have to a large extent created. In this volume will be found a very representative collection of tables now in ordinary use, both by statisticians and scientists who cope with their own statistical problems, and some that are new. Some of the better known ones, such as those of "t" and "z", are now considerably extended. Noteworthy additions are the tables of the Variance Ratio (known in America as "F"), the various transformation tables, including Bliss' Probits for the transformation of the Sigmoid Dosage Mortality Curve to a straight line, and those for use in the calculation of orthogonal polynomials. A novel departure is the inclusion of sets of Latin squares, which will be of value to those whose work calls frequently for this type of experimental design. Finally, there are the usual mathematical tables, such as logarithms, squares, square roots, reciprocals, factorials, natural sines and tangents. Those to whom calculating machines are still a luxury to be dreamt of only, will appreciate the sympathetic care with which these last tables have been assembled. Indeed, throughout, the book shows clearly that these tables have been regularly used by the authors and that personal experience has dictated the mode of presentation. To complete the book there are six pages of random numbers and a very comprehensive collection of mathematical constants, weights and measures. It goes without saying that a book of this nature can be used only by those who have at least some knowledge of the methods of statistical analysis involved. Nevertheless, in their introduction, the authors have done much to assist the

user by clear explanations of the scope of the tables and by examples of some of the uses to which they may be put. Altogether a book of which both the authors and printers may well be congratulated, and for which the world of biological research may well say "Thank you"!

T.N.H.

PLANT INJECTION FOR DIAGNOSIS AND CURATIVE PURPOSES. By W. A. ROACH. Foreword by Prof. V. H. BLACKMAN, Sc.D., F.R.S. (Technical Communication 10 of the Imperial Bureau of Horticulture and Plantation Crops, East Malling, Kent, England, 1938, pp. 78, plates 2, text figures 47, bibl. 162. 5s.)

Man has toyed for centuries with the idea of feeding his fruit trees direct instead of by more orthodox but slower methods, and in the hey-day of the Italian Renaissance we find Leonardo da Vinci noting that if it is desired to produce poisoned fruit, the injection of fruit trees offers a way. More recently various "tree doctors" have suggested that easy rejuvenation of orchards can be effected by the use of their own particular injection methods, but such methods have always been somewhat suspect, since obviously different conditions demand different remedies and the cure may prove worse than the disease.

It has remained for Dr. Roach to bring order out of chaos and show those of us who are attracted by the idea how to set about our investigations. He starts by giving a history of injection work in Europe and the U.S.A., paying particular attention to that published in recent years on the detection and cure of deficiency diseases. He notes also the successful use of injection for purely physiological purposes, as, for instance, of glucose into vines just before bud break, to ensure a good fruit set. He points out, moreover, how injection methods may be of great value in the study, not only of mineral deficiencies, but also of rootstock influence and of the effect of the chemical constituents of a fruit on its storage capacity.

Recent cases of successful injection cited include the following: diagnosis of incipient chlorosis of the iron shortage type at East Malling; diagnosis and cure of a copper deficiency dieback disease in apples in Western Australia; determination of the cause and cure of boron deficiency diseases of apples in Canada and New Zealand; increase in vigour and freedom from insect pests in apple trees injected with solutions of various chemical solutions in England.

His own experiments have been in progress for seven years on material which has ranged from the strawberry plant to fully grown apple and plum trees, and most of the present communication is devoted to a consideration of the methods used. It is for this clear and illustrated account that horticulturists will be particularly grateful. He describes in detail, with the help of text

figures and lists of tools, the methods used for the injection of particular parts of standing trees and other plants, varying in size from a single interveinal area of a leaf to whole main branches, noting, moreover, the type of problem for which each particular method is best suited. He shows how in the most delicate methods—the injection of leaves and their comparison with neighbouring untreated leaves or parts of leaves—enables a rapid diagnosis to be made of the mineral deficiency from which a plant or tree is suffering. He describes how whole trees may be injected for experimental purposes or even for economic reasons in the commercial orchard.

In short, he shows us how to carry out the operations advocated and how to avoid those many pitfalls which await the over-zealous, although he is far from suggesting that all his methods are perfect. He invites criticism and the experience of others in the same field.

BOOKS RECEIVED FOR REVIEW

PLANT FORM AND FUNCTION. By F. E. Fritsch and E. J. Salisbury. (G. Bell & Sons, Ltd., London. 17s. 6d.)

STATISTICAL METHODS FOR RESEARCH WORKERS. By R. A. Fisher. (7th Edition. Oliver & Boyd Ltd., Edinburgh. 15s.)

RESEARCH AND STATISTICAL METHODOLOGY BOOKS AND REVIEWS 1933-1938. Edited by Oscar Krisen Buros. (Rutgers University Press, New Brunswick, U.S.A. $1.25.)